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ANALYSIS OF DISSOLVED OXYGEN IN NATURAL AND WASTE WATERS

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ANALYSIS OF DISSOLVED OXYGEN IN NATURAL AND WASTE WATERS

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ABSTRACT

A comprehensive discussion of the various methods of analysis for dissolved oxygen in natural and waste waters is presented. Special emphasis is given to pointing out the applicability and limitations of each method. Several titrimetric methods and micro techniques are described. Manometric, gas-exchange, gas chromatographic, radiometric, and polarographic techniques and galvanic and membrane electrodes are among the instrumental methods discussed. In situ measurements of the activity of molecular oxygen are claimed to be more significant than concentration measurements. Examples of laboratory and field studies are given.

ANALYSIS OF DISSOLVED OXYGEN IN NATURAL AND WASTE WATERS

I. INTRODUCTION

Depending upon the intended use of the water, the presence of dissolved oxygen (DO) in water may be a blessing or a curse. Many industrial users desire water with no DO to inhibit corrosion, but those who are concerned with control of water pollution strive to maintain maximum solubility of atmospheric oxygen in water.

In water quality characterization, the analysis for DO is extremely important. Oxygen levels in streams are directly dependent on the physical, chemical, biochemical, and biological activities occurring in the water body; therefore, the analysis for DO is used as a primary indicator in pollution abatement and is a vital parameter in waste treatment process control.

DO measurements are often deceptively easy, but they can be exasperatingly difficult when high accuracy is required. Furthermore, there is not a general agreement as to the interpretation of most DO measurements. It is important, however, to recognize that most DO measurements are made because they represent an index of some biological or chemical activity. These being important variables, the effects of other extraneous environmental parameters on DO must therefore be ascertained, for they are the controlling factors on useful discrimination.

The main objective of this report is to present to the scientists and engineers dealing with water problems a concise, critical, and comprehensive, but not exhaustive, exposition on the modern aspects of analytical chemistry of DO. Emphasis is given to the theory and applicability of the various methods of analyzing DO in natural and waste waters.

Another objective of this report is that it will serve as a source of information for water chemists and for sanitary engineers in industry, government, and universities in the solution of routine and research problems in water and waste treatment. The report as a whole is intended to be a presentation of our present knowledge of the theory and practice of DO analyses and of the ways in which these procedures are applied to specific natural waters and industrial wastes in field use, analysis in situ, and continuous monitoring systems.

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II. PHYSICOCHEMICAL CHARACTERISTICS OF DISSOLVED OXYGEN

ORIGIN AND PROPERTIES

Oxygen is by far the most abundant element in nature. In the free state, molecular oxygen occurs mainly in the atmosphere and hydrosphere. Air is approximately 20 percent oxygen by volume; i.e., for every 100 molecules of air, approximately 20 are oxygen. On a weight basis, air is 21 percent oxygen; for every 100 grams (g) of air, approximately 21 are oxygen.

Surface waters get their oxygen supply mainly from the atmosphere and from the photosynthesis of green aquatic flora. Photosynthetic oxygen production by the aquatic flora may result in an oxygen oversaturation of bodies of water.

The solubility of atmospheric oxygen in water depends primarily on pressure, temperature, and salt content.

Effects of Pressure

Atmospheric oxygen solubility in water follows closely the laws of ideal gases, as shown in Figure 1, and can be calculated from Henry's Law.*

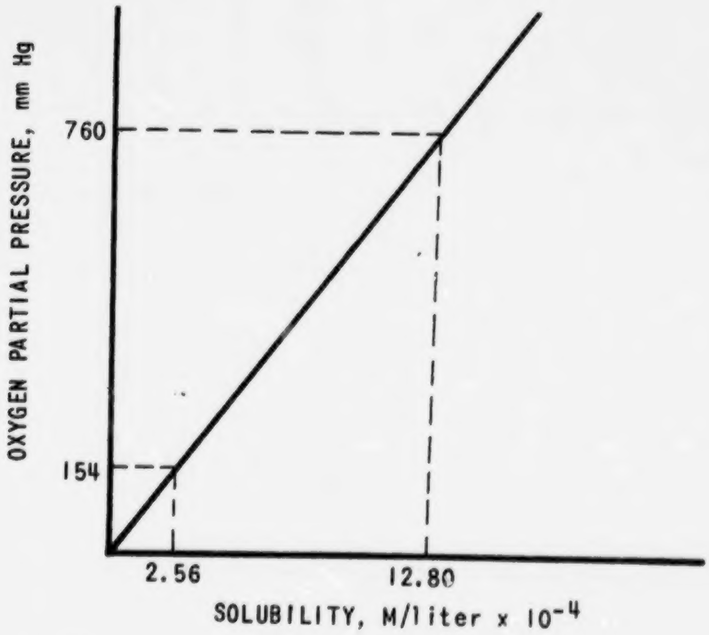

Figure 1. Solubility of oxygen in water at 25°C.

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*The solubility of molecular oxygen in water follows Henry's Law up to 4 atmospheres of oxygen.

$$P = HC \tag{1}$$

where: P = Partial pressure of oxygen = 154 mm Hg at sea level

H = Henry's Law constant = 5.98×10^5 mm Hg/M/liter

C = Oxygen concentration, M/liter.

Oxygen is a sparingly soluble gas in water; for example, at 25°C its solubility is 2.56×10^{-4} moles (M) per liter or 8.18 milligrams (mg) per liter at an oxygen partial pressure of 154 millimeters of mercury (mm Hg), whereas at 760 mm Hg the solubility is 12.80×10^{-4} M/liter, or 40.48 mg/liter.

The effect of hydrostatic pressure on the solubility of molecular oxygen in oceans and lakes, is highly significant. A thermodynamic analysis[54, 92] of this point involves the assumption that air at the surface of the ocean is in equilibrium with air dissolved in the surface water. In turn, air in the surface liquid layer is assumed to be in equilibrium with air dissolved in the next adjacent lower water layer and so on. This equilibrium exists down to any depth, h (h is measured positively in downward direction). Even though the solubility of a given gas (e.g., N_2 or O_2) at a deep level may be very different from that at the surface level, the chemical potential and the partial molal free energy will be the same for the solute at all levels. From this assumption, the change of the gas solubility with depth can be expressed as follows:

$$RT\, d(\ln C) = M\bar{g}\,(1 - \bar{v}\rho)\, dh \tag{2}$$

where: R = Gas constant = 8.3×10^7 ergs/M/degree

T = Absolute temperature, $^{\circ}K$

M = Molecular weight of solute, g/mole

$\bar{v}$ = Partial specific volume of solute, cm^3/g

ρ = Density of solution, g/cm^3

h = Depth, cm

$\bar{g}$ = Acceleration = 980 cm/sec^2

On the integration of equation (2) from the surface of the water to any depth, h, we obtain

$$RT \ln (C_{depth}/C_{surface}) = M\bar{g}\,(1 - \bar{v}\rho)\, h \tag{3}$$

Calculations from equation (3) indicate that as far as air is concerned, the solubility of oxygen remains practically constant with increasing depth. This is contrary to nitrogen, which decreases in solubility with depth. Although the solubility of oxygen is markedly affected by changes in temperature and salinity, the hydrostatic pressure seems to have no effect according to this model. For example, at T = 300°K, the solubility of oxygen is decreased by somewhat less than 0.1 percent at 1,000 meters below the surface.

Effects of Temperature

As with most gases, the solubility of oxygen in water decreases with increasing temperature. At 1 atmosphere (atm) of air, oxygen solubility in water at 0°C is 14.74 mg/liter, whereas at 35°C it is 7.03 mg/liter. This effect of temperature was found to conform to the following equation.[80, 102]

$$C = \alpha + \beta_1 T_c + \beta_2 T_c^2 + \beta_3 T_c^3 \qquad (4)$$

where: T_c = Temperature, °C

α, β_1, β_2, and β_3 = Constants.

An easier formula for the interpolation of DO saturation values at various temperatures is:

$$\frac{d(\ln C)}{d(1/T)} = \frac{\Delta H}{R} \qquad (5)$$

where: T = Absolute temperature, °K

ΔH = Heat of solution, cal/M

R = Gas constant = 1.99 cal/M/degree.

Although the heat of solution, ΔH, is not independent of temperature, it normally varies slightly so that use of log C and 1/T as variables provides a linear relationship. Integration of equation (5) gives

$$\log \frac{C_2}{C_1} = \frac{\Delta H}{2.3R} \left(\frac{1}{T_1} - \frac{1}{T_2}\right) \qquad (6)$$

Salt Content

Oxygen solubility in water is usually decreased by the addition of other solutes, particularly electrolytes. The extent of this "salting-out" effect varies considerably with different salts (Figure 2).

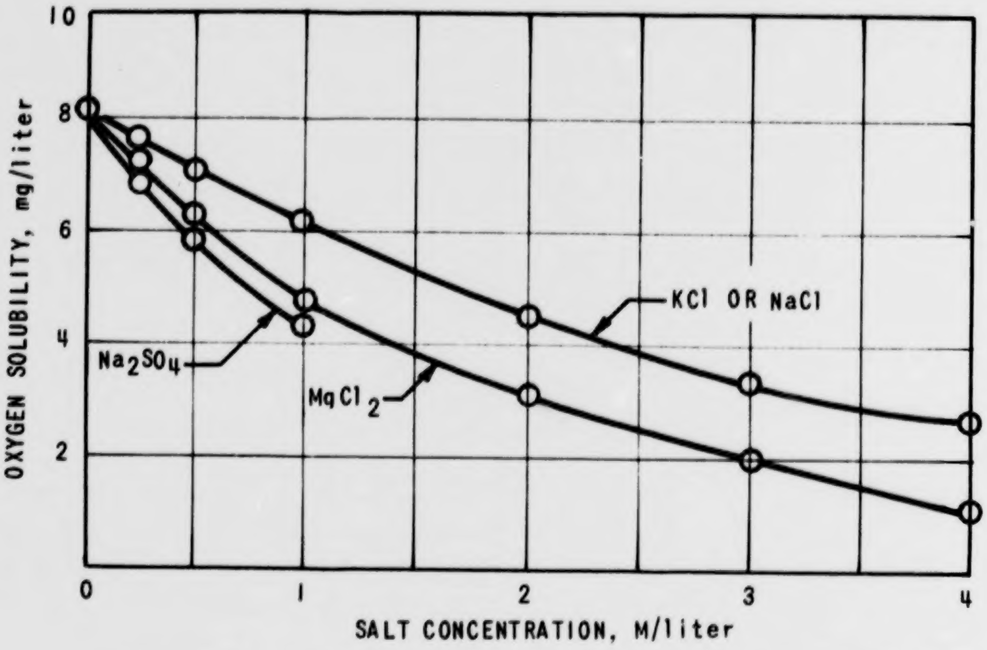

Figure 2. Oxygen solubility at various salt concentrations.

This effect may be better illustrated by considering the activity coefficient of molecular oxygen in aqueous systems "γ" defined as follows:

$$a = \gamma C \tag{7}$$

where: a = Activity, M/liter

γ = Activity coefficient

C = Concentration, M/liter.

The dependence of the activity coefficient* on the ionic strength of the salt solution is:

$$\ln \gamma = K_s I \tag{8}$$

where: K_s = Salting-out coefficient

I = Ionic strength.

Ionic strength can be defined as follows:

$$I = 1/2 \sum_i C_i Z_i^2 \tag{9}$$

where: C_i = Salt concentration of the "ith" component, M/liter

Z_i = Ionic charge of the "ith" component.

---

*The thermodynamic significance of the activity coefficient is discussed in Appendix E.

Accordingly, in distilled water the ionic strength, I, is zero, and by substitution in equation (8) the activity coefficient of the DO will be equal to unity. Under these conditions the activity of DO will be equal to the concentration. This is evident by substituting $\gamma = 1$ in equation (7).

In order to express the solubility of molecular oxygen as a function of the concentration of a given salt, the following equation is suggested:

$$\ln S = \ln S_o - K_s C_s \tag{10}$$

where: S = Oxygen solubility, mg/liter

S_o = Oxygen solubility in distilled water, mg/liter

C_s = Salt concentration, mg/liter

K_s = Constant.

The semilogarithmic plot of equation (10) gives a straight line with a negative slope equal to $-K_S$. It is important to understand that equation (10) is applicable only to electrolyte solutions of a single salt, whereas equation (8) can be used with solutions containing mixtures of salts.

SAMPLING

Great care is required in collecting water samples for the DO analysis. Methods of sampling are highly dependent on the source to be sampled and to a certain extent on the method of analysis. The sample must not remain in contact with air, nor be agitated, since either condition would cause a change in its gaseous content. Samples from any depth in streams, lakes, or reservoirs or samples from boiler waters need special precautions to eliminate changes in pressure and temperature. Procedures and equipment have been developed for sampling waters under pressure, (e.g., boiler waters), and unconfined waters (e.g., streams, rivers, and reservoirs). Sampling procedures and the equipment needed are described in American Society of Testing Material, Special Technical Publication No. 148-F[8] and Standard Methods for Analysis of Water and Wastewater.[105]

The time lag between taking a sample and performing the analysis is also of great significance. The longer the lag, the greater the chance that the DO content will change because of chemical or biological activity in the test solution.

Errors associated with the sampling of surface waters can be minimized by analyzing for DO in situ. Such measurements provide a more realistic value of the oxygen content in the natural environment.

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III. METHODS OF ANALYSIS

Each of the many methods of analysis for DO in aqueous systems utilizes a specific, basic, physicochemical property of molecular oxygen. For example, in the Winkler Test and in colorimetric methods the oxidizing property of oxygen is utilized, whereas voltammetric methods depend on the rate of diffusion of molecular oxygen. Other methods are based on the determination of the thermal conductivity or the paramagnetic properties of the oxygen molecules.

TITRIMETRIC METHODS

Winkler Test* and Modifications

The basic method for DO determination in natural waters is the Winkler Test, which was originally published about 75 years ago. It probably possesses most of the attributes of basic soundness and sensitivity. Improved by variations in equipment and technique, and aided by instrumentation, this test is still the basis for the majority of the analytical methods for analysis for DO. The reported[105] precision of the standard Winkler Test is given as a standard deviation of ± 0. 1 mg/liter.

Before using the Winkler Test for a DO determination in natural waters or wastes, the analyst must consider the effect of oxidizing or reducing materials in the sample. The presence of certain oxidizing agents liberates iodine from iodides (positive interference), and the presence of certain reducing agents reduces iodine to iodides (negative interference). Reducing compounds may also inhibit the oxidation of the manganous ion. Certain organic compounds were found to interfere with the Winkler Test in a different way. Surface-active agents were found to hinder the settling of the hydrated manganic oxide floc and partially obscure the end point of the iodometric titration with a starch indicator.

Several modifications of the Winkler Test are used to minimize the effect of interfering materials. Some of these methods are described below.

Alsterberg Modification - The most common interference is that caused by nitrites, commonly present in polluted waters and wastes. The nitrite ion reacts with iodides as follows:

$$2NO_2^- + 2I^- + 4H \longrightarrow I_2 + N_2O_2 + 2H_2O \qquad (11)$$

$$N_2O_2 + 1/2\ O_2 + H_2O \longrightarrow 2NO_2^- + 2H^+ \qquad (12)$$

---

*See Appendix A for the chemistry of the Winkler Test.

The nitrites formed in reaction (11) oxidize more iodide ions to free iodine, thus a cyclic reaction, which leads to high, erroneous results, is established. Also, the presence of nitrites in the test solution makes it impossible to obtain a permanent end point. As soon as the blue color of the starch-iodine complex disappears, the nitrites formed in reaction (11) react with more iodide to form iodine, and the blue color of the indicator appears again.

The Alsterberg modification[3] utilizes sodium azide, which reduces the nitrites in the following manner,

$$NaN_3 + H^+ \longrightarrow HN_3 + Na^+ \qquad (13)$$

$$HN_3 + NO_2^- + H^+ \longrightarrow N_2 + N_2O + H_2O \qquad (14)$$

Rideal-Stewart Modification - This modification is proposed to overcome the interferences caused by a wide variety of reducing agents.[95] The sample is pretreated with potassium permanganate solution under acid conditions to oxidize reducing agents as shown by the following:

$$5NO_2^- + MnO_4 + 6H^+ \longrightarrow 5NO_3 + 3H_2O + 2Mn^{2+} \qquad (15)$$

$$5Fe^{2+} + MnO_4^- + 8H^+ \longrightarrow 5Fe^{3+} + 4H_2O + Mn^{2+} \qquad (16)$$

$$\text{Aldehydes} + MnO_4 + H^+ \longrightarrow \text{Acids} + Mn^{2+} + H_2O \qquad (17)$$

Under the best condition the nitrates formed in reaction (15) do not oxidize the iodide ions; also if the ferric iron formed in reaction (16) is below 10 mg/liter, it does not interfere with the test. Above that level, however, ferric ions can be precipitated as the salt FeF_3,

$$Fe^{3+} + 3F^- \longrightarrow FeF_3 \qquad (18)$$

Aldehydes and other organic compounds that exert an iodine demand are oxidized to their respective acids.

After the permanganate has oxidized the reducing compounds present, the excess is destroyed by potassium oxalate,

$$(COO)_2^- + MnO_4^- + 8H^+ \longrightarrow 2CO_2 + Mn^{2+} + 4H_2O \qquad (19)$$

The difficulty in manipulation of the Rideal-Stewart Modification usually results in low accuracy and precision.

Alkaline Hypochlorite Modification - The alkaline hypochlorite modification[114] was designed to overcome interferences caused by sulfur compounds, e.g., wastes of the sulfite pulp industry, which may contain sulfites, thiosulfates, polythionates, etc. This procedure involves the pretreatment of the sample with alkaline hypochlorite to oxidize the sulfur compounds to sulfates and sulfur. Excess hypochlorite is destroyed by potassium iodide, and the iodine formed is titrated by sodium thiosulfate.

Again, this modification is difficult to perform, and the results obtained are of low accuracy.[84]

Alum Flocculation Modification - Suspended solids in water samples may consume certain quantities of iodine during the Winkler Test.[96] This interference may be removed by flocculation (with alum and ammonium hydroxide) and settling of the solids prior to the regular Winkler Test. This modification also is reported to be of low accuracy.[84]

Copper Sulfate - Sulfamic Acid Flocculation - For samples from biological treatment units, e.g., activated sludge,[96, 97] the addition of copper sulfate to the sample prior to the test precipitates the biological floc. Sulfamic acid is also added to inhibit biological activity. After the floc settles, an aliquot from the supernatant liquor is siphoned and analyzed for its DO content by the usual Winkler Test. The copper sulfate and the sulfamic acid are combined into a single solution in practice. This modification is also considered to be of low accuracy.[84]

Short Theriault Modification - In the Winkler Test, at a high pH value organic matter may be hydrolized by the addition of the alkali-iodide solution, which create an "oxygen demand, " or at low pH value acidification may cause the oxidation of the organic matter by the free iodine. The Theriault modification speeds up the test by reducing the mixing time so that the interaction of organic compounds with iodine or iodides in the test solution is minimized.

Pomeroy-Kirschman-Alsterberg Modification - The Pomeroy-Kirschman-Alsterberg modification[88] is designed for DO determination of supersaturated water samples. The method utilizes saturated solutions of the alkali-azide-iodide reagent. It is claimed that using concentrated reagents will also provide for high organic concentrations in the sample.

Lederer Modification - Lederer[57] claimed that the use of cerous instead of manganous salts eliminates interferences from organic substances, nitrites, and iron.

Erdery and Szabadvary Modification - In this modification the water sample is treated with sodium hydroxide and ferrous ammonium sulfate.[33] Oxygen in the test solution oxidizes the ferrous ions to ferric, which are titrated with ascorbic acid using 4-amino-4'-methoxydiphenylamine as an oxidation reduction indicator. The authors claim that this procedure minimizes the effect of interferences by oxidizing agents.

Yakimets and Shabanova Modification - It was suggested that the addition of NH_2SO_3H together with the manganous salt will eliminate interference from nitrites.[127]

Potter and White Modification - To remove ferrous iron, especially when analyzing for low DO concentrations, an ion exchange column was used.[90] Precautions were taken to deoxygenate the resin bed before introducing the sample. The main difficulty in the use of ion exchange resins is the slow attainment of an oxygen equilibrium between the sample and the water held by the resin.

Iodine Difference Method - This method[89, 91] employs the addition of a known small quantity of iodine solution to two portions of the water, one of which is a control. The standard Winkler Test is then performed on the other. The presence of reducing substances reacts with iodine in both the sample and the control; therefore, the difference between the titration of the sample and the control gives the iodine equivalent of DO in the sample. A correction for the effect of any reducing compounds may then be made. It is important to maintain the same experimental conditions and time of contact for both the sample and control after the addition of the iodine solution.

Reversed-Reagent Method - This procedure has been used to compensate for interferences present in the water sample as well as in the reagents.[59] Two water samples, A and B, of equal volume are used. Sample A is treated with the usual Winkler Test. The order of addition of reagents to sample B, the blank, is reversed, i.e., KOH and KI are added first, followed by H_2SO_4 and $MnSO_4$. In this case the DO is not being "fixed" by the manganous salt. Because the blank is initially made alkaline, allowance is made for substances that interfere specifically in an alkaline medium. Interfering substances in the samples as well as in the reagents react in both samples. Accordingly, the difference between the sample and the blank (A minus B) represents the oxygen in sample plus the oxygen in the reagents.

If reducing substances such as sulfite are present in greater amounts than oxidants, it is possible to have a "negative" blank. In this case identical quantities of iodine or iodate are added to both sample and blank to provide an excess of iodine after acidification.

The reversed-reagents method is commonly used, but it does not correct for interferences caused by ferrous iron. This leads to low results by forming ferrous hydroxide, which reacts with oxygen in the blank. The ferric ions formed do not produce an equivalent amount of iodine upon acidification.

O-tolidine Method - After the DO has oxidized the divalent manganese in the conventional Winkler Test, the solution then may be acidified with chlorine-free concentrated hydrochloric acid.[20, 36, 75, 76, 103] This reaction results in the liberation of an equivalent quantity of chlorine according to the following reaction:

$$MnO_2 \cdot H_2O + 4H^+ + 4Cl^- \longrightarrow Mn^{2+} + 3H_2O + 2Cl_2 \qquad (20)$$

The free chlorine then reacts with orthotolidine to form the characteristic yellow-colored holoquinone chloride. The intensity of the color of the holoquinone chloride is directly proportional to the oxygen content of the sample and is measured by comparison with standards, colored slides, or by the use of adsorption spectrometry. Recent studies* revealed that the chloride ion is not necessarily an intermediate in the production of chlorine, which does the oxidation of the orthotolidine. Colloidal manganese (IV) produced by air oxidation of manganese (II) is capable of reacting directly even in neutral solution with orthotolidine in the absence of chlorides. It is also important to note that air oxidation of manganese (II) does not produce manganese (IV), but rather a mixed oxide.[79]

The O-tolidine modification is popular because of its simplicity and the availability of the permanent color discs for residual chlorine.[45, 59, 60, 112]

3:3 - dimethylnaphthidine (DMN) Method - After the regular Winkler reagents, sodium acetate, and DMN[12] are added to the test solution, color develops. Its intensity is directly proportional to the original DO. This method has been used mainly in Europe to analyze for DO in boiler feed waters.[108]

Direct Titration Method - In this modification the DO oxidizes the manganous salt to a higher valency state. This step is followed by a direct titration with a standard ferrous ammonium sulfate solution and ferrous o-phenanthroline as an indicator.[118]

A similar approach has been adapted for boiler waters in which the DO is reacted with an excess of an acid chromous solution. The residual chromous ions are titrated with standard potassium biniodate.[107]

Iodometric Titration

The critical step in the Winkler DO determination is the quantitative estimation of the released free iodine. This may be done either by titrimetric or colorimetric techniques. Iodine titration in the standard Winkler method[105] is carried out according to the following reaction:

$$I_2 + 2S_2O_3^{2-} \longrightarrow S_4O_6^{2-} + 2I^- \qquad (21)$$

The end point is usually detected visually by means of starch as an indicator. The complex compound formed by the interaction of iodine with starch possesses a characteristic blue color,† which can be used to detect the presence of molecular iodine as low as 10^{-5}M.

---

*J.D. Johnson. School of Public Health, University of North Carolina, Chapel Hill, N.C., private communications.

†Amylose, which is the straight chain fraction abundant in potato starch, forms the blue color with iodine. The other major fractions of starch are branched in structure, and they form a red-purple color complex with iodine.

There are, however, two main sources of error in the direct titration of free iodine. An error may occur because of iodine's volatility (low solubility in water, 1.33×10^{-3} M at 20°C), usually encountered in solutions of low iodide concentrations. Loss of gaseous iodine also occurs through changes in temperature or on agitation.

A second possible source of error is air-oxidation of iodide to iodine. This reaction is highly dependent on the pH of the test solution, the rate of oxidation increasing rapidly with increasing acidity. Also, the oxidation of iodide to iodine is catalyzed to a certain extent by a variety of metal ions, e.g., Cu^{2+}, and is subject to photochemical action.

Because of the likelihood of errors during a direct titration for free iodine, alternative titrimetric techniques may be employed, as discussed in the following section.

Potentiometric Method - The potentiometric method involves the measurement of the oxidation-reduction potential during the titration process. This measurement gives a quantitative expression of the strength of the oxidation-reduction system according to the following equation:

$$E_m = E_o + \frac{RT}{nF} \ln \frac{(\text{Oxidant})}{(\text{Reductant})} \tag{22}$$

where: E_m = Measured potential, v

E_o = Standard potential, v

R = Gas constant = 8.31 joules/M/degree

T = Absolute temperature, °K

n = Number of electrons transferred in oxidation-reduction process

F = The Faraday.

The apparatus used for a potentiometric titration is quite simple and is essentially composed of a titration cell (Figure 3) and a device for the determination of the cell voltage. The titration cell contains an indicator electrode and a reference electrode. A commercial calomel electrode (industrial type) from pH meter manufacturers is convenient to use as the reference electrode. The solution is stirred continuously by a magnetic stirrer during the course of the titration. For routine control analysis, potentiometric titrators are used to automatically record the titration curve or automatically stop the addition of titrant at the end point.

A typical oxidation-reduction potentiometric titration curve is shown in Figure 4. The end point of a titration is characterized by the inflection point in the curve. On plotting the first derivative of the measured

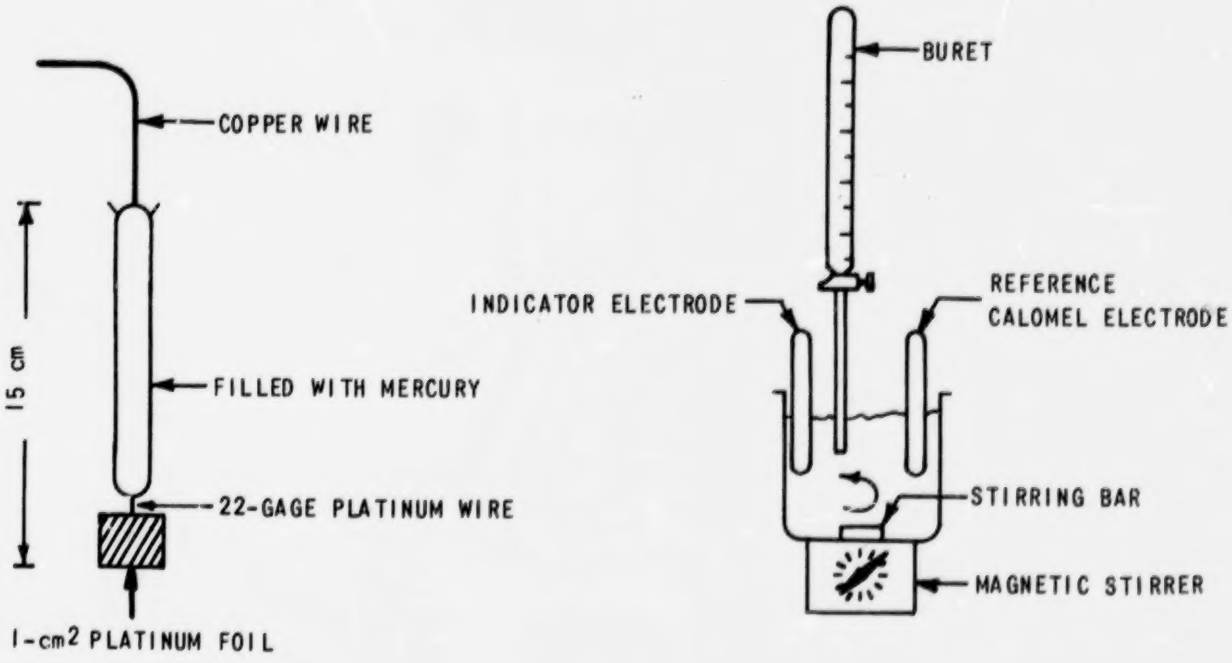

COPPER WIRE
15 cm
FILLED WITH MERCURY
22-GAGE PLATINUM WIRE
1-cm2 PLATINUM FOIL
BURET
INDICATOR ELECTRODE
REFERENCE CALOMEL ELECTRODE
STIRRING BAR
MAGNETIC STIRRER

Figure 3. Equipment for potentiometric titration.

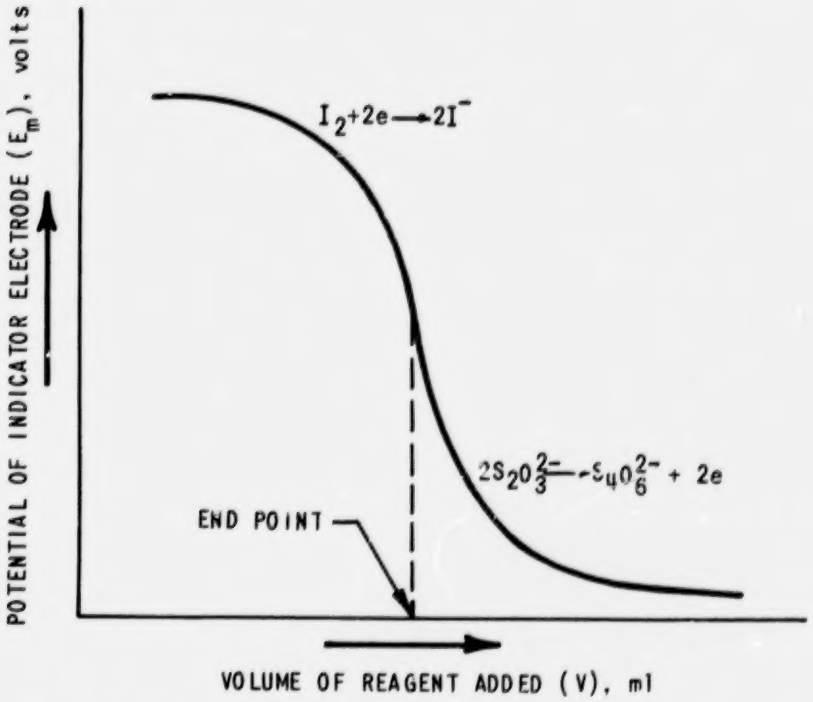


Figure 4. Potentiometric titration of iodine.

potential, E_m, and the volume of reagent added, dE_m/dV, versus the volume of reagent, the end point is characterized by the peak in the curve. (Figure 5).

The precision of the potentiometric titration technique is reported to be about ± 0.001 mg/liter of iodine.[89]

"Dead-Stop" End-Point Method - This special type of amperometric technique, noted for its simplicity, is probably the most precise and sensitive method for titrating iodine with thiosulfate. A small potential

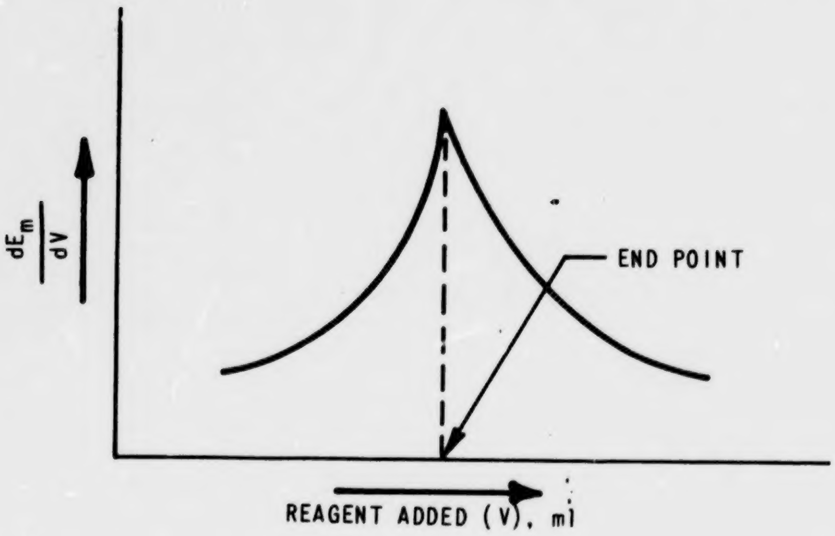


Figure 5. Derivative potentiometry titration curve.

difference (15 to 400 mv, depending on the sensitivity required) is set up between two smooth platinum electrodes in the solution to be titrated (Figure 6). The diffusion current is measured during the course of the titration. No attempt is made to control the potential of either electrode during titration, only the potential difference is controlled. The end point is indicated by the disappearance of the current flowing in the cell as shown in Figure 7.

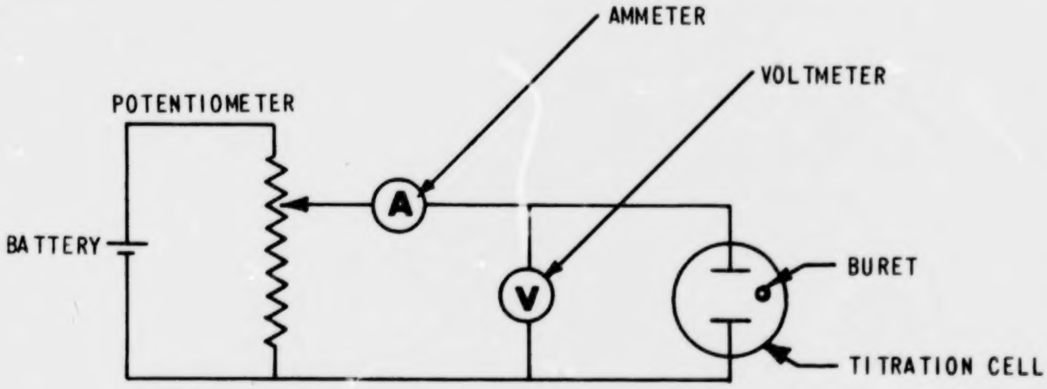


Figure 6. Dead-stop end-point circuit.

The course of the titration is as follows: As long as free iodine remains in the solution, the chief electrode reaction, under the influence of the applied voltage, will be the oxidation of iodide to iodine at the anode and the reverse process at the cathode. At the end point and when the free iodine is removed, the iodine to iodide reaction can no longer occur and the cell current will come to a "dead stop." Since the thiosulfate/tetrathionate reaction is highly irreversible and proceeds at only a minute rate under the influence of the applied voltage, no detectable current is observed at and beyond the end point. Ordinarily, the end point is so easily detected that there is no need for a graphical estimation of its position. By using sensitive current-measuring devices

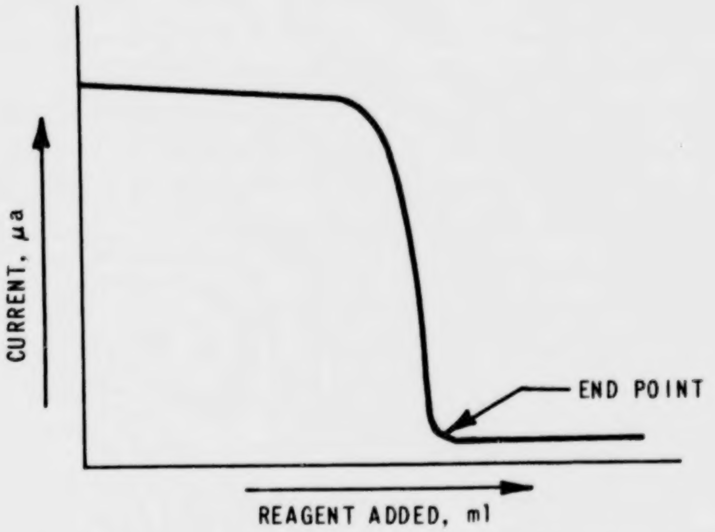


Figure 7. Dead-stop end-point titration curve.

the end point can be established to an accuracy of ± 0.01 microgram iodine in a 100-milliliter sample, or 1 part in 10 billion.

Coulometric Titration Method - Coulometric titrations are those in which the titrant (or reagent) is electrically generated at the surface of electrodes immersed in solution. Coulometric titrations follow Faraday's Law.* expressed mathematically as follows:

$$G = \frac{qM}{nF} \tag{23}$$

where: G = Weight of substance (iodine), g

M = Molecular weight of substance, g/mole

q = Number of coulombs passed

n = Number of equivalents per molecular weight (or the number of electrons in the half-cell reaction)

F = The Faraday.

Most coulometric titrations are done at constant current to simplify the instrumentation and to produce an answer directly proportional to time as shown by the following equation.

$$q = i \int_0^t dt = it \tag{24}$$

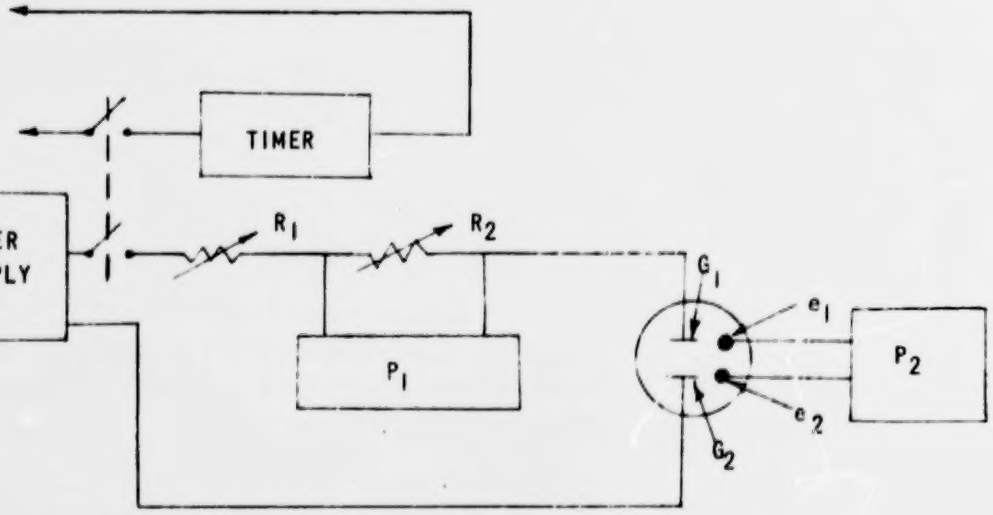
where: t = Time, sec

i = Current, amp $\times 10^{-6}$.

---

*Faraday's Law: The passage of 1 Faraday (96,500 coulombs) of electricity will oxidize or reduce 1 gram-equivalent of a substance.

A typical constant-current coulometer is illustrated in Figure 8. The procedure for coulometric titration of DO consists of the addition of $MnSO_4$, $KOH+KI$, H_2SO_4, and an excess of $Na_2S_2O_3$ standard solution successively to the test solution.[44] The iodine formed electrolytically reacts with the residual thiosulfate in solution. The electrolytic current is held constant by varying the potential, and the equivalent point is conveniently detected by the dead-stop end-point method. The concentration of DO is calculated from equations (23) and (24). Since i is constant, by substitution of time of electrolysis, t, in equation (24), q (the number of coulombs) is determined. By substitution of q in equation (23) the weight, g, of DO in the sample can be calculated.



R_1, R_2 = RESISTORS
P_1 = POTENTIOMETER FOR CURRENT MEASUREMENT
P_2 = POTENTIOMETER FOR END POINT DETECTION
e_1, e_2 = MONITORING ELECTRODES
G_1 = GENERATING ELECTRODE
G_2 = AUXILIARY ELECTRODE

Figure 8. Constant-current coulometric circuit.

The coulometric titration method is very accurate, to within 0.02 µg/liter. It has a distinct advantage in that since the titrant is generated in solution, errors caused by contact with air, as in conventional titrations, are eliminated.

General Remarks -

1. Samples for the Winkler Test should be treated immediately with the reagents to "fix" the DO. They may be stored before, but not after, the liberation of the iodine.[40]

2. Sulfuric acid can be replaced by hydrochloric acid, provided the latter is free from ferric iron (common impurity), since this readily oxidizes iodide to iodine. Sulfuric acid may contain traces of nitrogen oxide impurities, which can be removed by fuming the acid.

3. Free iodine (see Appendix A, equation A-3) is volatile, and titration (of sample or standards) should be done rapidly and preferably with stoppered Erlenmeyer flasks.

4. Standardization of the thiosulfate solution can be done with a standard potassium dichromate, arsenious oxide, or potassium biniodate solution. The biniodate solution is by far the most convenient and most reliable reagent. Iodine solutions are not recommended as a primary standard because of volatility. The reaction of potassium dichromate with iodide ions is not instantaneous, and hydriodic acid in the presence of chromic salts may be air-oxidized.

5. Instead of starch, disodium glycollate, which may be added at the beginning of the titration, may be used as an end-point indicator. Using either of these indicators, however, has been found to lead to results on the low side.[56]

6. The effect of the pH of the test solution[40] after the addition of the acid on the accuracy of the Winkler Test is illustrated in Figure 9. These results clearly indicate that enough acid must be added to bring the pH below 1.7.

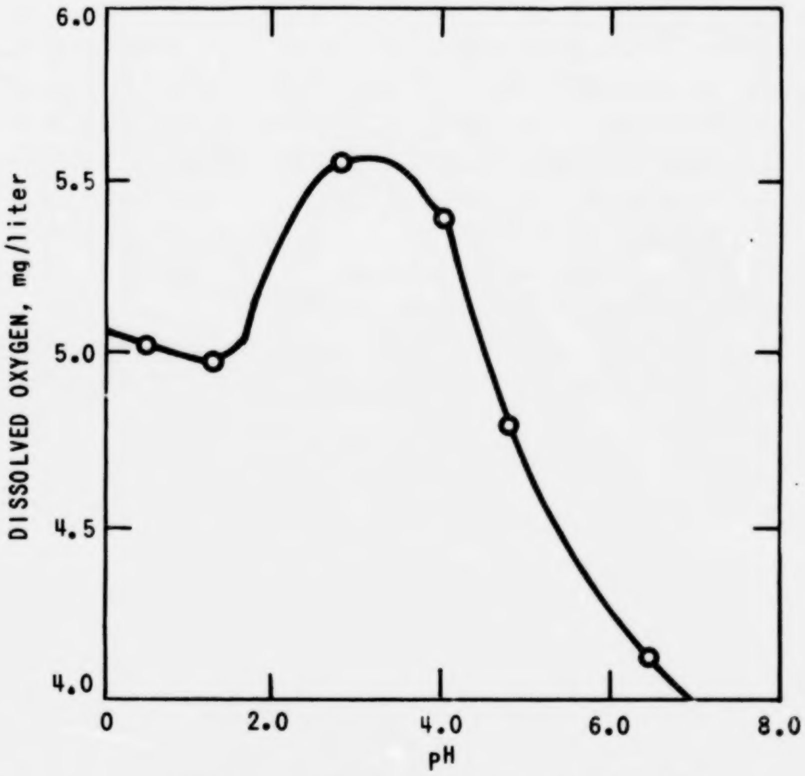


Figure 9. Effect of pH on Winkler Test.

The high DO values in the pH range of 1.7 to 3.7 are explained by the slow rate of dissolution of $MnO_2 \cdot H_2O$ as given by equation (A-3). Under these conditions, atmospheric oxygen causes the oxidation of iodide to iodine, which results in a higher consumption of thiosulfate. Above pH 5.0 the reaction given by equation (A-3) is no longer quantitative because of insufficient hydrogen ions.

Other Modifications

Micro-Winkler Method - The Winkler method as described in Standard Methods[106] is not suitable for DO determinations in small volumes of sample, nor in biological systems where repeated DO measurements with small volume changes are required. A modified micro-method[122] has been developed; it uses 1 to 2 milliliters of water and has an accuracy of 1 to 2 percent. The apparatus consists of a "syringe pipet," Figure 10, in which both the water sample and the reagents are allowed to react. The liberated iodine is then expelled for titration. The procedure is given in Appendix B.

Technique Variations - Variations in technique have been proposed in an attempt to improve the accuracy and the sensitivity of the Winkler Test.

In one method[128] the sensitivity is increased threefold by withdrawing most of the supernatant liquor from the settled manganic hydroxide. If two-thirds of the total liquid volume has been removed, on acidification, a threefold enrichment of the free iodine is attained. In another suggested procedure,[7] the liberated iodine in the Winkler Test is removed by several successive extractions with carbon tetrachloride. Starting with a 1-liter water sample, the iodine may be concentrated in a final volume of about 30 milliliters and determined colorimetrically or by titration.

Discussion

In spite of the various modifications proposed to reduce or minimize the interfering effects of certain compounds on the Winkler Test, most of these modifications are complicated and of no practical significance. The exceptions are the iodine difference method and the Alsterberg modification involving the decomposition of nitrites by sodium azide.

Apart from the effect of interferences, the Winkler Test itself has limitations:

1. Being a titrimetric method, it is not considered a suitable field method.
2. It cannot be adapted for DO determination in situ.
3. The Winkler Test cannot be adapted to continuous monitoring for water quality control purposes.
4. The Winkler Test is essentially a destructive test, and the sample used cannot be returned to its source. This is only of significance in laboratory experiments, where it is necessary to know the DO of a

small volume of solution over a long period of time.

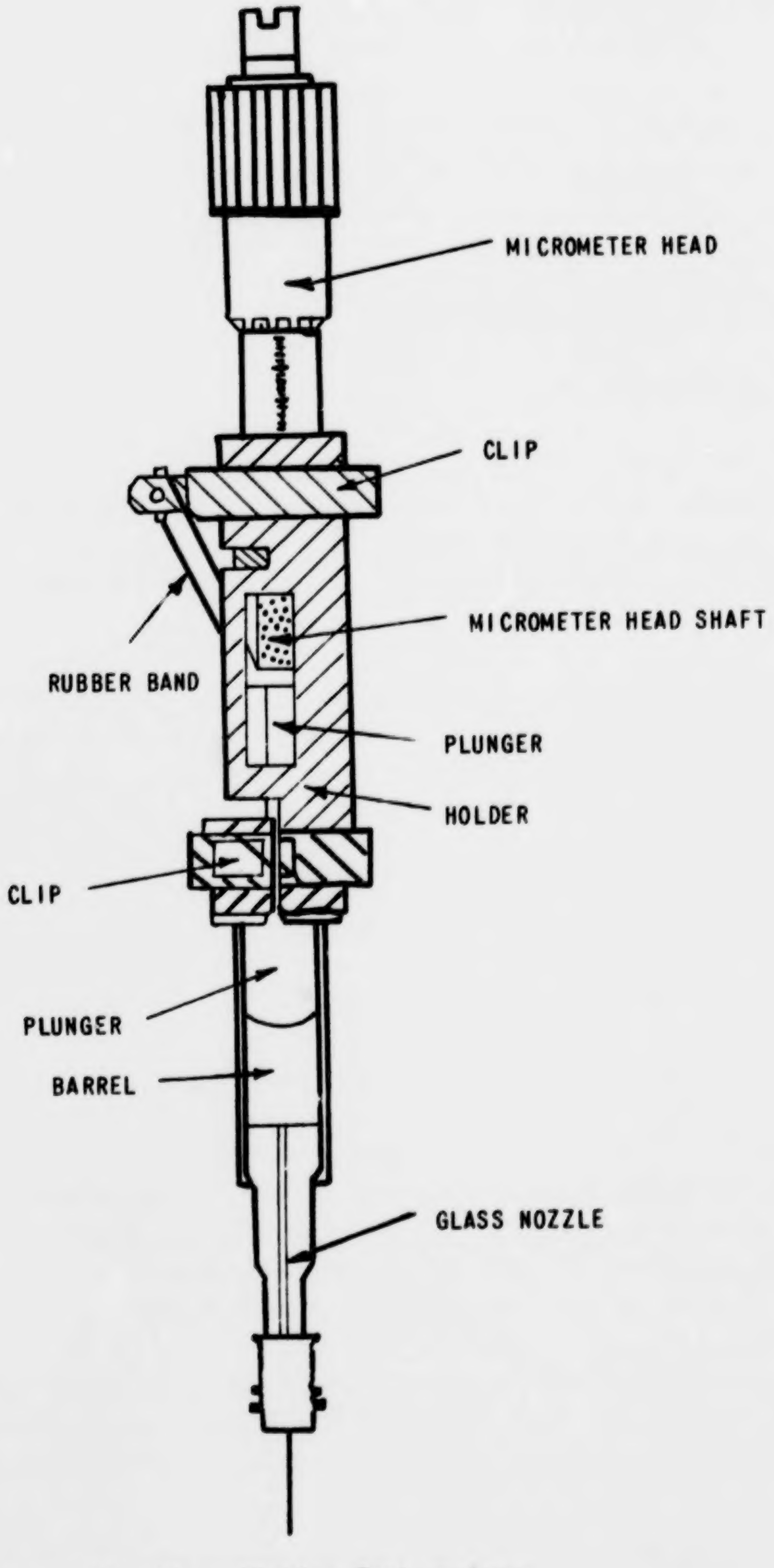

MICROMETER HEAD
CLIP
RUBBER BAND
MICROMETER HEAD SHAFT
PLUNGER
HOLDER
CLIP
PLUNGER
BARREL
GLASS NOZZLE

Figure 10. Micro-Winkler Test syringe.

COLORIMETRIC METHODS

Colorimetric Determination of Iodine

The intensity of the blue color of the starch-iodine complex can be measured photometrically.[10, 87] Careful control of temperature and other parameters make it relatively difficult, however.

The liberated iodine can be directly determined with high accuracy[7, 9] by absorption spectrometry at a wavelength of 450 millimicrons (mμ).

Free iodine can be extracted from the aqueous solution into a small volume of carbon tetrachloride and the color intensity determined photometrically by disc comparators.[7]

Direct Determination

Direct colorimetric methods of analysis are based on the interaction of molecular oxygen with a chromogenic agent (an oxidation-reduction indicator) to give a color change. One of the most commonly used indicators for the detection of oxygen in solution is methylene blue; others are indigo carmine and safranin T.

In the presence of DO a methylene blue solution exhibits a blue color, and in the absence of DO it is colorless. This indicator has been used in the relative stability test of sewage effluents, an expression of the time required for the decolorization of a water or waste sample.[105]

Quantitative colorimetric determinations of DO can be made, however, by means of indigo carmine dyes. Indigo carmine in the reduced state reacts with oxygen to give a color change through orange, red, purple, blue, and finally a blue green in the completely oxidized form.*

The method has been used with a high degree of accuracy in the range of 0-100 ppb for boiler waters.[1, 21, 64, 65] A special sampling assembly[64, 65] enables the addition of the reduced indigo carmine to the water without oxygen contamination. The resulting color can be compared with temporary working standards made of colored solutions of inorganic salts or with colored glass disc standards.[1] In the range of 0 to 25 ppb, color changes of 2 ppb can be detected visually.

Other oxidation-reduction dyes have also been used for the colorimetric analysis for DO, such as the leuco form of safranin T,† reportedly giving the same precision and accuracy[2] as indigo carmine.

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*Indigo carmine solution is prepared by dissolving the dye in a 1:1 solution of glycerol in water; to this solution is added 1% KOH for pH control and 1% glucose as the reducing agent.

†The safranin reagent is a 0.1% solution in 7:3:10 water, 20% ammonium hydroxide, and ethyl alcohol.

Discussion

It was reported that metal ions such as nickel, cupric, or zinc ions up to 1,000 ppb; ferric ions at 3,000 ppb; and sulfite up to 12 ppb do not interfere. Ferrous ions, however, were found to interfere, and it was suggested that these be removed by an ion exchange column.[21]

Oxidizing and reducing agents commonly present in waste water interfere with the direct colorimetric methods.[77] In relatively clean surface waters these methods can be used successfully.

The direct colorimetric methods have the following limitations:

1. For accurate analysis, the reduced indigo carmine must be added without exposing the water sample to atmospheric oxygen. A special sampling assembly is, therefore, required. This complication results in difficulties in the field.

2. Direct colorimetric methods have not been adapted for DO determination in situ.

3. They cannot be easily adapted for continuous monitoring purposes.

4. The test is destructive, i.e., the sample cannot be returned to its source.

ION-EXCHANGE TITRIMETRIC METHOD

In this ingenious method a cation exchange resin is converted to the ferrous form with 0.1N ferrous ammonium sulfate.[45] The sample to be analyzed is then passed through the prepared resin. DO in the test solution oxidizes an equivalent quantity of ferrous iron to ferric. The total iron, both ferrous and ferric, is eluted out of the column by 6N sulfuric acid. After titration of the unoxidized ferrous ions in the eluate with potassium permanganate solution, the ferric ions are calculated. The concentration of DO in the test solution is equivalent to the ferric ions in the eluate.

This method is only applicable to high-purity water. Oxidizing agents, complexing agents, and solids in the test solution interfere seriously with the test.

GAS EXCHANGE METHOD

The main difficulty encountered in the determination of DO in natural and waste waters is the effect of interfering substances present in the sample. One way to avoid this effect is to "pull out" oxygen molecules from the test solution and determine their quantity in the gas phase. This is the basic principle of the gas-exchange oxygen analyzers. The method used is to exchange oxygen in the test solution with a carrier gas and subsequently, to measure the oxygen in the carrier gas. A typical oxygen analyzer of this type is composed of two parts, the gas exchange unit and the oxygen-detecting unit.

Gas Exchange Unit

A carrier gas must be inert (e.g., nitrogen or hydrogen) so that it does not react with oxygen under test conditions or activate the sensing element in the oxygen-detection unit. The gas exchange unit is usually composed of an aspirator followed by a water-gas separator, as shown in Figure 11. In the aspirator unit, the water sample with the carrier gas is forced through a nozzle at about 50 pounds pressure. Intimate mixture of the gas and water results in the transfer of oxygen from the aqueous phase to the gas stream (Henry's Law). The extent of the gas transfer in this process depends on the efficiency of the gas exchange unit. It is doubtful, however, that an instantaneous equilibrium partition of oxygen between the incoming sample and the carrier gas will take place; therefore, water and gas flow rates must be kept uniform to maintain a certain degree of gas exchange.[113]

The carrier gas leaving the exchange unit is passed through the oxygen-detecting unit and then is either vented to the atmosphere[113] or recirculated through the exchange unit.[28, 59, 60, 67] In the latter technique, after the carrier gas has been circulated a few times, equilibrium conditions are attained. For all practical purposes, at equilibrium conditions, the oxygen in the solution is assumed to be completely replaced by the carrier gas.

Oxygen-Detecting Unit

Three methods are commonly used to determine the oxygen content in the carrier gas.

Oxygen-Sensitive Galvanic Cell - Oxygen-sensitive galvanic cells are composed of a relatively basic metal anode, e.g., zinc, cadmium, or lead; a relatively noble cathode, e.g., gold, silver, or nickel (or carbon); and a highly conductive electrolyte solution, e.g., potassium hydroxide. The carrier gas containing oxygen is bubbled through the electrolyte solution by means of diffusers. The oxygen goes into solution and is subsequently depolarized at the cathode; an equivalent amount of current is thus generated.[41, 53] The generated current, directly proportional to the oxygen content in the original water or waste solution, is usually measured by a microammeter calibrated in terms of oxygen.

An example of the reactions in an oxygen-sensitive galvanic cell made of a zinc-carbon couple in a 1$\underline{M}$ potassium hydroxide solution is as follows:

Anodic reaction (Zinc electrode)

$$Zn \longrightarrow Zn^{2+} + 2e \qquad (25)$$

$$Zn^{2+} + 4\,OH^- \longrightarrow ZnO_2^{2-} + 2\,H_2O \qquad (26)$$

GPO 826-743-3

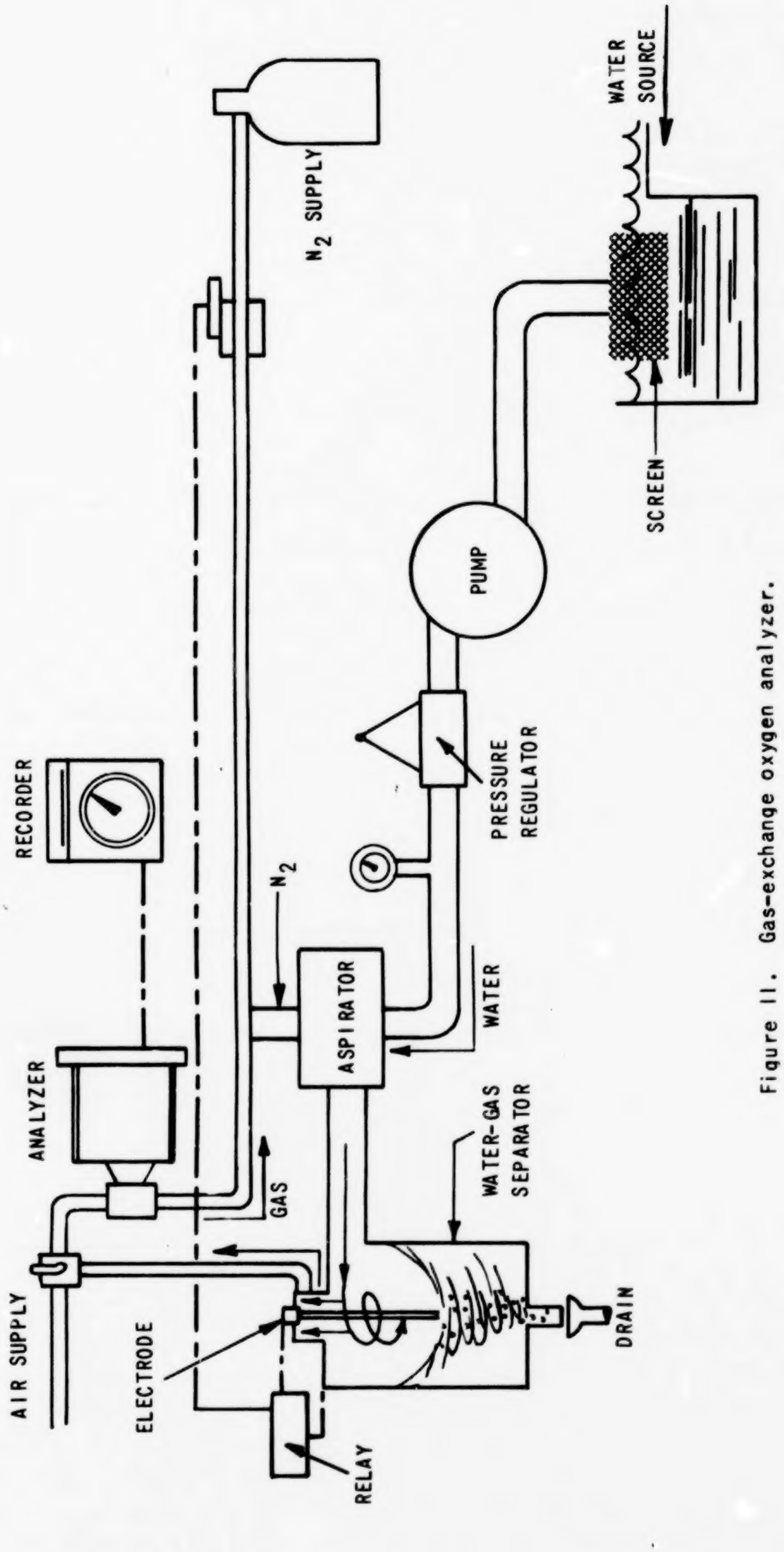

AIR SUPPLY
ELECTRODE
RELAY
DRAIN
WATER-GAS SEPARATOR
GAS
ANALYZER
ASPIRATOR
WATER
N_2
RECORDER
PRESSURE REGULATOR
PUMP
SCREEN
N_2 SUPPLY
WATER SOURCE

Figure 11. Gas-exchange oxygen analyzer.

Cathodic reaction (Carbon electrode)

$$1/2\ O_2 + H_2O + 2e \longrightarrow 2\ OH^- \qquad (27)$$

Overall reaction

$$Zn + 2\ OH^- + 1/2\ O_2 \longrightarrow ZnO_2^{2-} + H_2O \qquad (28)$$

Paramagnetic Method - Magnetic properties are not confined only to ferromagnetic materials. All substances are influenced by a magnetic field, although to a lesser degree than a material like iron. Substances attracted to a magnetic field are called "paramagnetic," and those repelled are called "diamagnetic." All liquids and gases fall into one or the other of these classes. Oxygen is unique among gases in that it possesses relatively high paramagnetic properties, whereas most gases are diamagnetic (Figure 12). Accordingly, it is possible to analyze for oxygen by means of its magnetic susceptibility.[13]

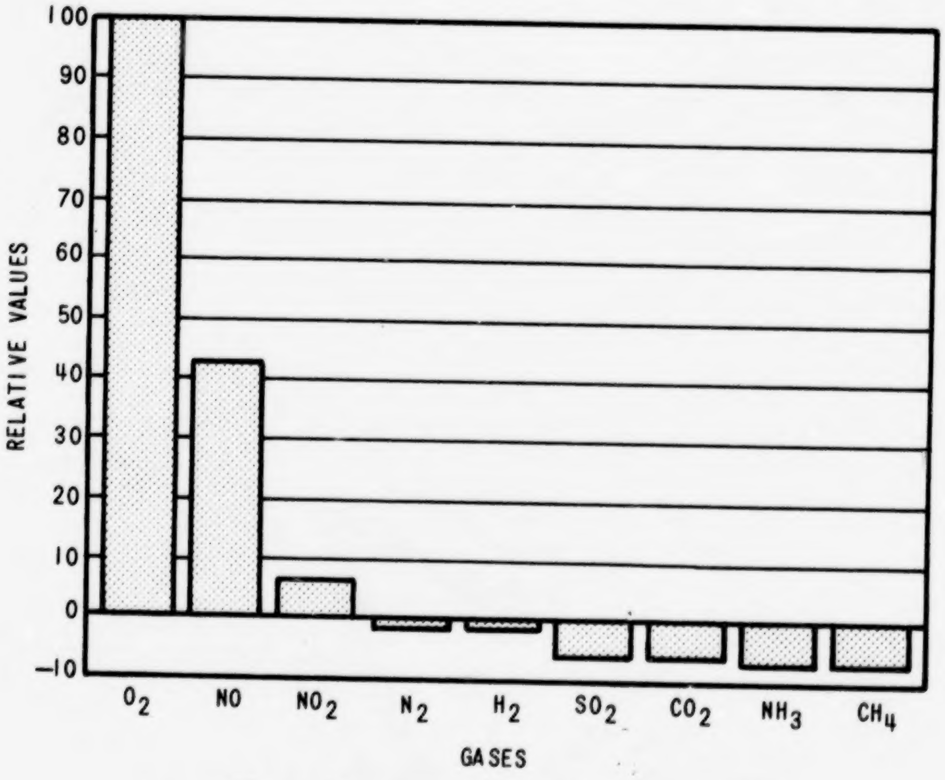


Figure 12. Magnetic properties of gases.

A carrier gas leaving the exchange unit passes through the oxygen-detecting unit containing a magnetic field in the vicinity of a hot spiral wire,[41] as shown in Figure 13. Upon being heated, molecular oxygen loses its magnetic properties and cooler oxygen molecules attracted into the magnetic field replace those that have been heated. This results in a circulation of gas in the vicinity of the spiral wire. The change in the resistance of the spiral wire caused by the flow of the cooler gas is proportional to the amount of oxygen present in the stream of the carrier gas, which, in turn, is equal to the DO content of the test solution. In another method for magnetically determining the oxygen content in the

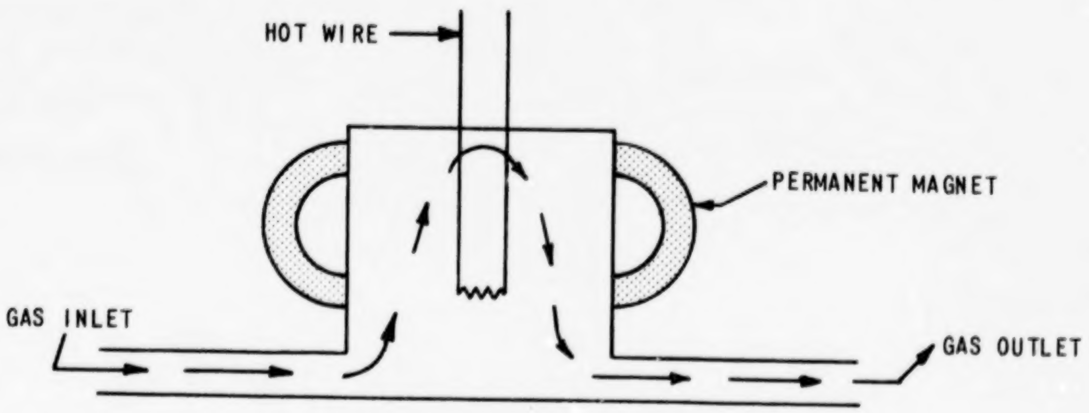


Figure 13. Paramagnetic analyzer cell.

carrier gas, a small glass dumbbell is suspended on a taut quartz fiber in a nonuniform magnetic field (Figure 14). When oxygen is absent, the magnetic force exactly balances the torque of the quartz fiber and the dumbbell remains stationary. When the carrier gas containing oxygen is drawn into the detecting unit, the magnetic field is altered and the dumbbell rotates. The degree of rotation is proportional to the change in the magnetic force, which, in turn, is proportional to the oxygen concentration in the sample. By means of a small mirror attached to and rotating with the dumbbell, a beam of light is projected on a translucent scale, which is usually calibrated in terms of oxygen concentration.[15]

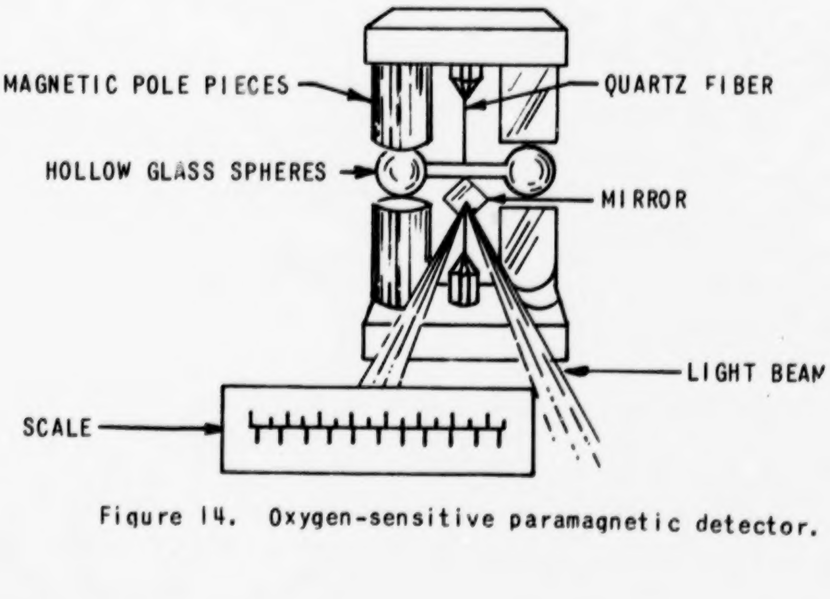


Figure 14. Oxygen-sensitive paramagnetic detector.

Thermal Conductivity Method - The thermal conductivity of a gas is defined as the quantity of heat transferred in unit time between two unit surfaces when they are a unit distance apart and the temperature difference between surfaces is 1°C.[27] The thermal conductivity of oxygen is usually determined in reference to the carrier gas (hydrogen or nitrogen).

Thermal-conductivity oxygen-detecting units contain a wire with a high temperature coefficient of resistance (e.g., platinum) stretched along the axis of a metal or glass cylinder and connected to the arm of a Wheatstone bridge circuit, as shown in Figure 15. When a current is passed through the bridge, the wire becomes heated. The equilibrium temperature of the wire depends, however, on the thermal conductivity

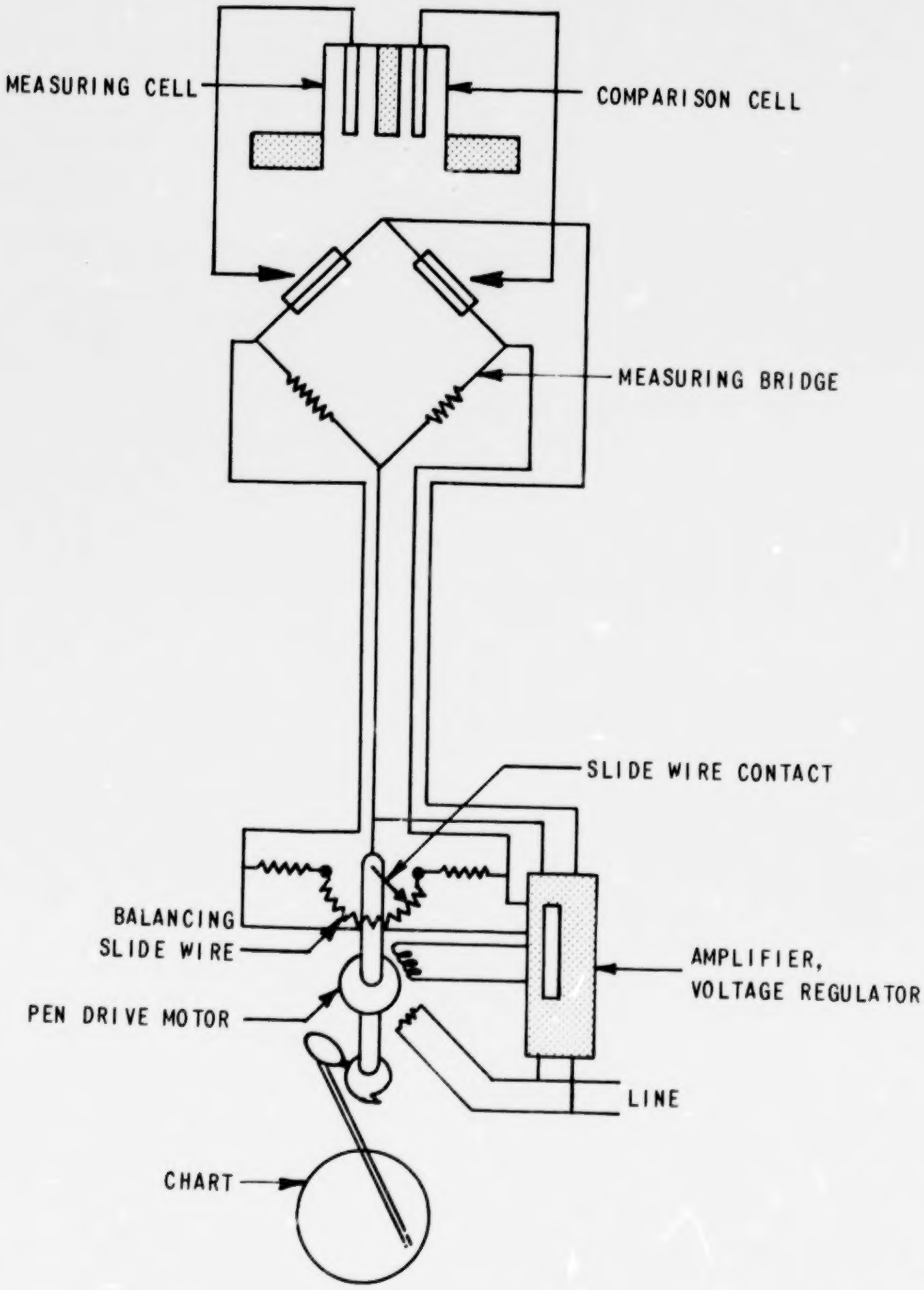

MEASURING CELL
COMPARISON CELL
MEASURING BRIDGE
SLIDE WIRE CONTACT
BALANCING SLIDE WIRE
AMPLIFIER, VOLTAGE REGULATOR
PEN DRIVE MOTOR
LINE
CHART

Figure 15. Schematic diagram of thermal conductivity oxygen analyzer.

of the surrounding gas. Thus, changes in the thermal conductivity of the carrier gas due to the oxygen exchange cause an imbalance in the bridge. The imbalance is measured by a galvanometer or potentiometer, which can be calibrated in terms of the DO content of the test solution.[42]

Discussion

Gas-exchange oxygen analyzers have been used for DO determinations in natural waters and wastes.[30, 47, 59, 60, 67] The main advantages of these systems are:

1. Analyzing for oxygen in the gas phase makes it possible to eliminate the interference of ionic and organic compounds commonly present in natural water and waste.
2. The units are adaptable to continuous monitoring of DO.
3. These units are suitable for field operations; e.g., they can be installed on the bank of a stream or on the side of an activated-sludge unit.

Gas-exchange oxygen analyzers, however, have the following drawbacks:

1. These oxygen analyzers are unable to determine DO in situ since they are not submersible.
2. The instruments are neither portable nor rugged to the extent that they can be mounted in a boat for a river survey.
3. These analyzers are not suitable for use on discrete samples in the laboratory.

The accuracy of the gas-exchange oxygen analyzers usually varies from ± 2 to ± 5 percent of full-scale reading. Thus for a range of 0 to 5 mg/liter full-scale deflection, the accuracy is between ± 0.1 to ± 0.25 mg/liter. The precision of such instruments is about ± 1 percent of full-scale deflection.

GAS CHROMATOGRAPHIC METHOD

The method, as the name implies, depends on the determination of molecular oxygen in a gaseous stream.[123] The dissolved gases are stripped from the test solution with an inert gas, followed by chromatographic separation and subsequent detection of its components. The stripping is done in a gas exchange unit consisting of multiple spinning discs rotating at high speed, partially immersed in the water sample as shown in Figure 16. This unit is able to purge the water sample of its gas content in a single pass through, at inert gas / water flow ratios of 10 to 1 through 1 to 2.

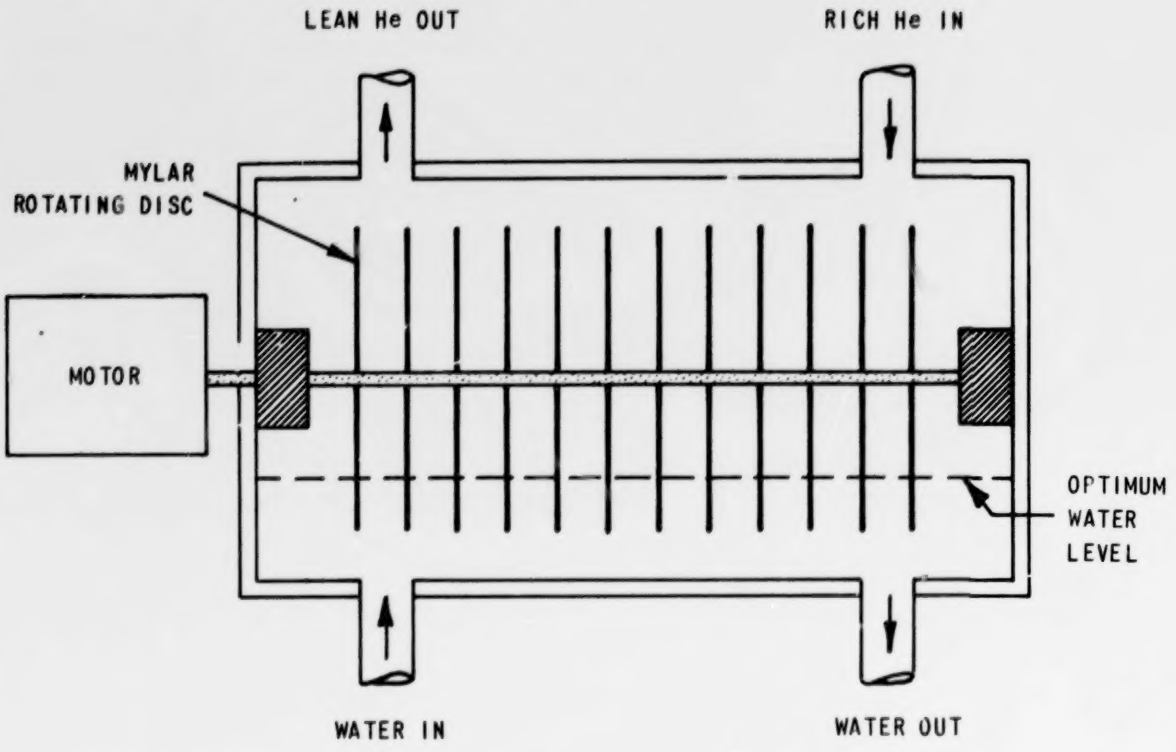


Figure 16. Gas exchange unit.

The gas stream from the exchange unit is then passed through two gas chromatographic columns in series (Fisher Gas Partioner, Model 25) as shown by the block diagram in Figure 17. After the chromatographic separation of the different components, the gas stream is passed through the detector cells (thermal conductivity cells) and the elution curve is recorded by a 10-millivolt full-scale recorder.

The instrument is calibrated in terms of peak height versus gas volume, e.g., millimeters peak height versus milliliters of O_2 per liter. Good results were obtained by using helium as the carrier gas. When argon was used, the gas partitioner could not separate it from oxygen. A suggested alternative procedure[123] involves the analysis of duplicate samples, one of which has had the oxygen removed by catalytic combustion or absorption before passing through the chromatographic columns.

Discussion

The gas chromatographic method offers the following advantages:

1. Like the previously discussed gas exchange method, the chromatographic method eliminates the effect of interferences commonly present by analyzing for oxygen in the gaseous phase.
2. The chromatographic method is an on-stream device that can be adapted for continuous monitoring in plant operations.
3. This method can be used for the analysis for all gases in the test solution (CO, CH_4, N_2, and CO_2) simultaneously with oxygen.

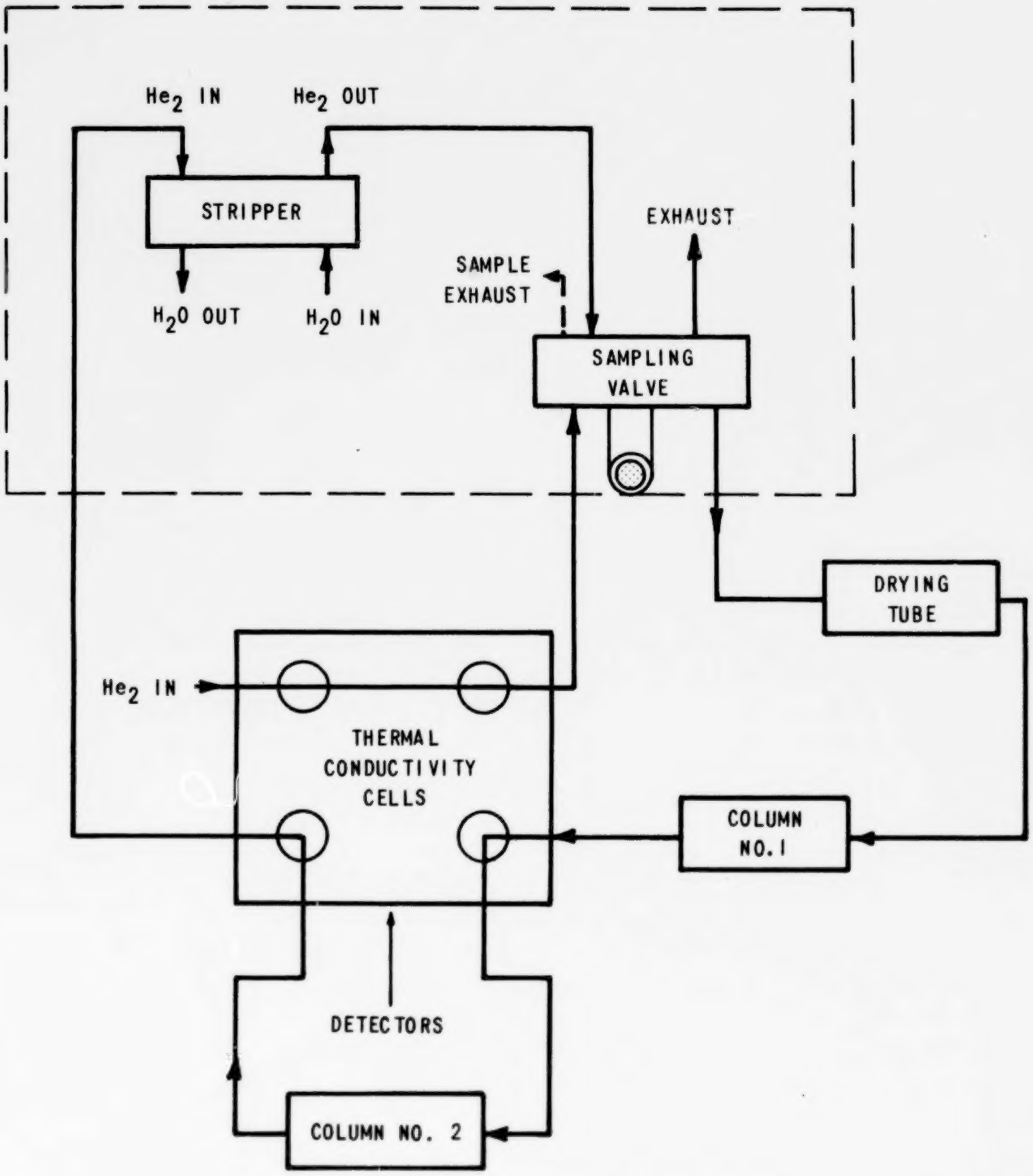

He_2 IN
He_2 OUT
STRIPPER
H_2O OUT
H_2O IN
EXHAUST
SAMPLE EXHAUST
SAMPLING VALVE
DRYING TUBE
He_2 IN
THERMAL CONDUCTIVITY CELLS
COLUMN NO. 1
DETECTORS
COLUMN NO. 2

Figure 17. Gas chromatographic analyzer.

The gas chromatographic method is, however, a relatively new method, and it has not been tried out in the field; therefore, an evaluation of its performance characteristics cannot be given. There are, however, certain inherent operational drawbacks, which may be summarized as follows:

1. It is a highly delicate, complex, instrument that needs specialized attention and constant regulation, e.g., the regulation of carrier gas and the water flow rates.

2. The method is unable to analyze for DO in situ.

3. The instrument lacks the ruggedness and portability required for field use.

GASOMETRIC METHOD

The gasometric method is a modified Van slyke gasometric procedure similar to that described by Theriault and McNamee[114] and recently modified by McKeown et al.[74]

The method consists of stripping all gases from a test solution under vacuum. The gases are then transferred to a separate reaction vessel, which contains the Winkler reagents. After shaking for 15 to 20 minutes, the contents of the reaction chamber are then titrated with thiosulfate, and the oxygen concentration is determined. Detailed descriptions of the gasometric method and the apparatus are given in Appendix C.

A detailed discussion on the effect of the sample volume, duration of stripping, and time of reaction has been investigated.[74] A correlation between the Winkler Test and the gasometric method in distilled water samples showed excellent agreement at DO levels greater than 1.0 mg/liter. At DO concentrations below 1.0 mg/liter slight turbulence caused by transferring the sample to the gasometric apparatus may result in considerable error. The accuracy of the gasometric method was determined to be ± 0.21 mg/liter.

Discussion

The gasometric method may be considered as a gas exchange method since the analysis for oxygen is done on the gaseous phase. Similar to gas exchange methods, the main advantage of this method is the elimination of the interference of ionic and nonvolatile organic compounds present in the test sample. This does not, however, exclude the effect of volatile species, e.g., H_2S, which interfere with the Winkler determination in the reactor vessel.

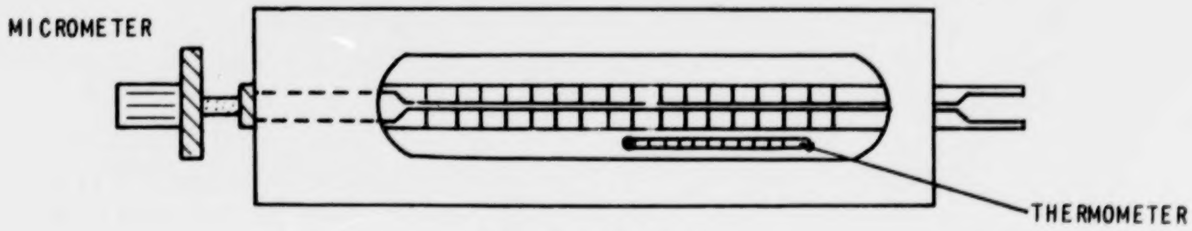
The main limitations of the gasometric method are:

1. The apparatus cannot be easily adapted to continuous monitoring of DO.

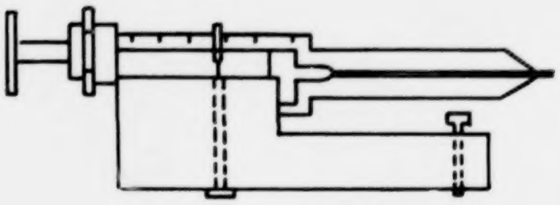
2. The gasometric method is strictly a laboratory procedure and cannot be considered as a field test.

MICRO-GASOMETRIC METHOD

With the micro-gasometric method DO can be determined in as small as 1-milliliter water sample.[100] The technique consists of stripping out the dissolved gases from a 1-milliliter sample in a specially designed pipet (Figure 18). The gases, in the form of a bubble, are then allowed to react with an alkali for the removal of carbon dioxide gas. A pyrogallol solution is next introduced into the syringe to absorb the oxygen gas. The change in the volume of the bubble is equal to the volume of DO in the sample. Details of the experimental procedure are given in Appendix C.



BUBBLE ANALYZER



SYRINGE PIPET

Figure 18. Microgasometric method equipment.

The precision of the micro-gasometric method was reported to be within ± 2 percent.[100]

Discussion

The main advantages of the micro-gasometric methods are:

1. Analysis for DO and nitrogen simultaneously is possible. With simple modifications in procedure, the carbon dioxide content can also be determined, but with much lower accuracy.
2. The technique can be used in the field.
3. Many interfering materials usually present in polluted waters have no effect.

The micro-gasometric method has the following limitations:

1. The method cannot be easily adapted for continuous analysis.
2. It cannot be used for DO analysis in situ.

RADIOMETRIC METHOD

The radiometric method of analysis is based on the quantitative oxidation of radioactive thallium-204 by the oxygen in the test solution.[94] Thallium-204 is primarily a beta emitter and has a half-life of 3.6 years; therefore, decay over several months does not greatly reduce the sensitivity of the technique.

The apparatus consists of a column of radioactive thallium metal electro-deposited on copper turnings, and two flow-type Geiger-Mueller counters (Figure 19).

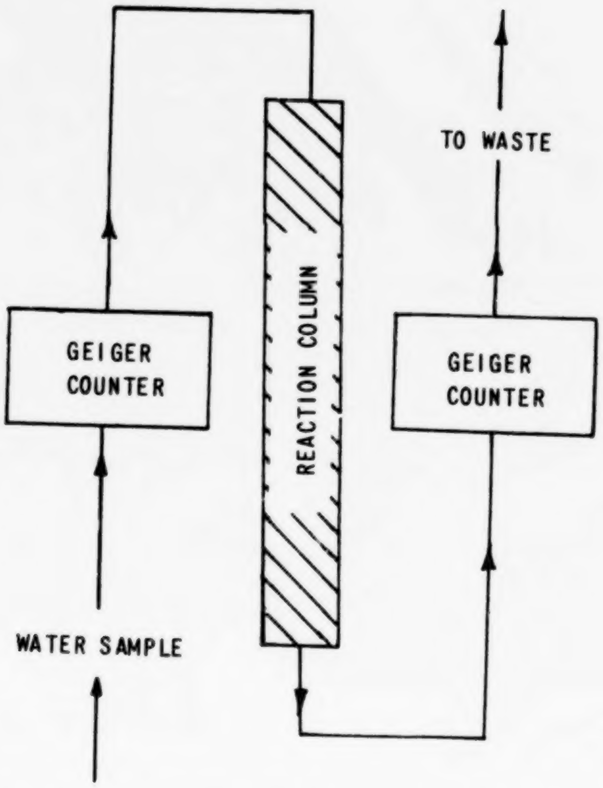


Figure 19. Radiometric method apparatus, employing thallium-204.

The technique involves passing the test solution by one of the Geiger-Mueller counters to detect any background beta-activity, then through the column where the following reaction occurs.

$$4\,Tl_{(s)} + O_{2\,(aq)} + 2H_2O_{(liq)} \longrightarrow 4\,Tl^{+}_{(aq)} + 4\,OH^{-}_{(aq)} \qquad (29)$$

The radioactive thallium ions in the effluent of the column are detected by the second Geiger-Mueller counter. One milligram of DO will liberate 25.6×10^{-3}g of ^{204}Tl. The counting rate is directly proportional to the DO concentration in the test solution.

Discussion

The radiometric method offers the following advantages:

1. The method is adaptable to continuous monitoring.
2. The instrumentation involved is not particularly complicated despite the complexity of the column preparation.

The radiometric method suffers from the following drawbacks:

1. The counting rate is not linear with the DO in seawater because of the electrolytic effect on thallium ion solubility.
2. The pH of the test solution has a marked effect on the rate of corrosion of thallium ions in the reaction column.
3. Oxidizing agents may be expected to produce a marked interference with this method at low pH values.
4. The sensitivity of this method using a column with a specific activity of 2.04 millicurie (mc) per gram of thallium is about 0.2 mg/liter. That is to say, a test solution containing 0.2 mg/liter produces a ^{204}Tl counting rate equal to the background counting rate of the detector. As a rule, because of the randomness of radioactive disintegrations, the precision of this method is ± 2 percent.

CONDUCTOMETRIC METHOD

Conductometric methods of analysis are based on the reaction of the DO of the test solution with compounds to form ionic species and increase proportionally the conductance of the test solution.

One method measures the conductivity of a flowing sample of water to which nitric oxide gas is added.[34] The DO oxidizes nitric oxide molecules to nitrite ions according to the following reaction.

$$4\,NO + O_2 + 2H_2O \longrightarrow 4H^+ + 4NO_2^- \qquad (30)$$

The increased ionic strength of the test solution results in an increase in electrical conductance directly proportional to the DO concentration.

A flow diagram of this instrument is shown in Figure 20. The water sample passes first into ion-exchange columns to eliminate interferences from bases and buffer salts. Then it is passed through a reference conductivity cell and through the reaction column, where it comes in contact with the nitric oxide gas. The reaction of molecular oxygen with nitric oxide goes to completion instantaneously.[34] The sample is then passed through the second conductivity cell. The change in conductivity is usually recorded in terms of the DO concentration.

In another approach, instead of using nitric acid gas, the water sample is passed into a column containing shavings of a solid reactant such as lead[124] or thallium metal.[126] The oxygen reacts with these metals to form their respective ions (anodic corrosion).

$$2\,Pb + O_2 + 2H_2O \longrightarrow 2Pb^{2+} + 4\,OH^- \quad (31)$$

$$\text{or}\quad 4\,Tl + O_2 + 2H_2O \longrightarrow 4\,Tl^+ + 4\,OH^- \quad (32)$$

The pH of the test solution is important since it influences the reactions.

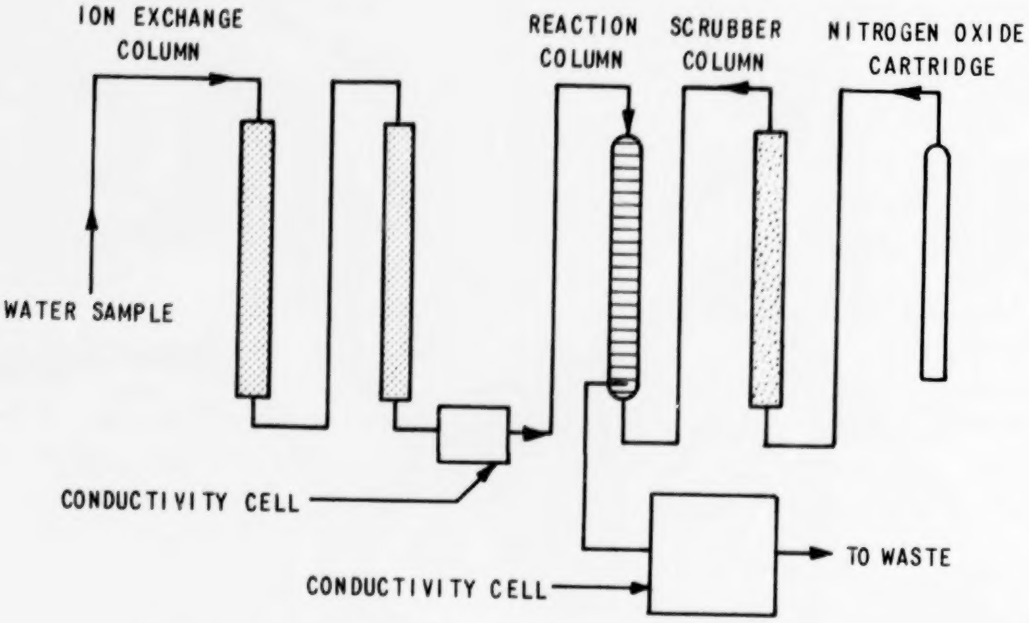

ION EXCHANGE COLUMN
REACTION COLUMN
SCRUBBER COLUMN
NITROGEN OXIDE CARTRIDGE
WATER SAMPLE
CONDUCTIVITY CELL
CONDUCTIVITY CELL
TO WASTE

Figure 20. Conductivity apparatus.

Since the equivalent conductance of thallous hydroxide is 253, 1 mg/liter of DO will produce an increase in conductivity of 32 micromhos per centimeter (μmhos/cm). It is important to note, however, that the background conductivity of different surface waters may vary from 100 to 1,000 μmhos/cm. To measure minute amounts of DO, the water is first pretreated, if necessary, with ion-exchange resins to insure a pH $\geq$ 7, reduce initial conductance of the sample, and eliminate carbonates and phosphates, which interfere with the measurements. After the water sample has passed over the Pb or Tl surface, the change in conductivity is recorded. This may be expressed in DO concentration units.

Discussion

Analysis by means of conductometric methods has been used successfully in boiler feed waters. These methods offer the following advantages:

1. Relatively high sensitivity of a few parts per billion.[126]
2. Adaptable for continuous monitoring.

On the other hand, conductometric methods have the following drawbacks:

1. Incapable of analyzing for DO in situ.

2. Pretreatment of the sample by passing it through ion-exchange columns probably causes a loss of DO because of the large reaction surfaces. This, in turn, may activate reactions between molecular oxygen and organic or inorganic substances in the water sample.

COULOMETRIC METHOD

For purposes of orientation, it is helpful to note that air saturated water at 25°C and under 750 mm Hg air pressure contains 8.18 mg/liter DO. This in terms of coulometric analysis is 0.1083 ampere. second/gram (amp. sec/g) of water, a rather large measurement.

Few coulometric methods of analysis for DO have been reported. Two of these[13, 44] are discussed here, and both utilize the principle of constant-current coulometry. In one method,[44] a deoxygenated solution of chromic ions is added to the water sample. During the test, chromous ions are generated electrolytically and are then oxidized by the oxygen in solution to the chromic state. The apparatus immediately detects any excess of chromous ions, marking the end of the titration. This method is sensitive to 0.03 mg/liter of DO and has an accuracy of ± 2 percent.

Another coulometric method[13] is based on the interaction between DO and an ammonium-copper complex $[Cu(NH_3)_2]^+$. This is followed by the reduction of the oxidized ammonium-copper complex $[Cu(NH_3)]^{2+}$ on a platinum cathode. The amount of current used is equivalent to the DO content in the test solution.

Recently, a constant-potential derivative coulometric system was used.[32] The working electrode was reported to be composed of tiny metal spheres packed in a ceramic tube. No field experience has been reported on this system.

Discussion

Coulometric methods of analysis for DO are only suitable for high-purity waters containing no interferences. Under these conditions, previously described coulometric methods are very sensitive and exhibit high accuracy.

VOLTAMMETRIC METHODS

Voltammetric (potential-current) methods depends on the electrolytic reduction of molecular oxygen at a polarized,* inert, sensing electrode to generate an electric current that is directly proportional to the oxygen content of the test solution. The electrode reaction is dependent on the nature of the electrode material, the pH of the solution, and the potential of the electrode-solution interface.

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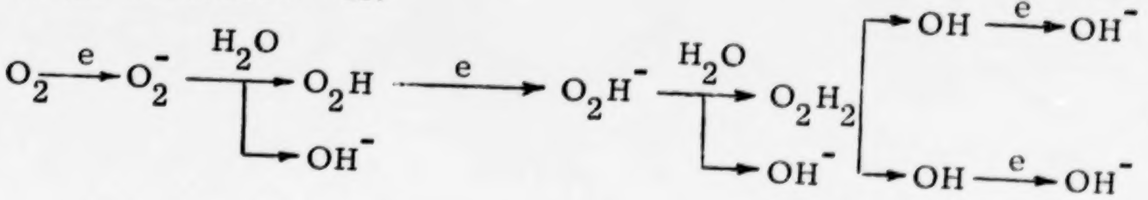
*A polarized electrode is one that is forced to a potential other than the equilibrium potential of the solution.

The mechanism of the electrolytic reduction of molecular oxygen is not yet well known. It is dependent on the type and the surface properties of the cathode; sorption and desorption; proton transfer; the split of an O-O bond; and, in the case of some metal electrodes, the growth and decay of transient oxide films. A schematic, simplified approach showing possible intermediates during the reduction of molecular oxygen on metal electrodes[34] is:



$O_2 \xrightarrow{e} O_2^-$
H_2O
O_2H
OH^-
$\xrightarrow{e} O_2H^-$
H_2O
O_2H_2
OH^-
$OH \xrightarrow{e} OH^-$
$OH \xrightarrow{e} OH^-$

(33)

$$O_2 + H_2O + 2e \longrightarrow OH^- + HO_2^- \qquad (34)$$

$$O_2 + 2H_2O + 4e \longrightarrow 4OH^- \qquad (35)$$

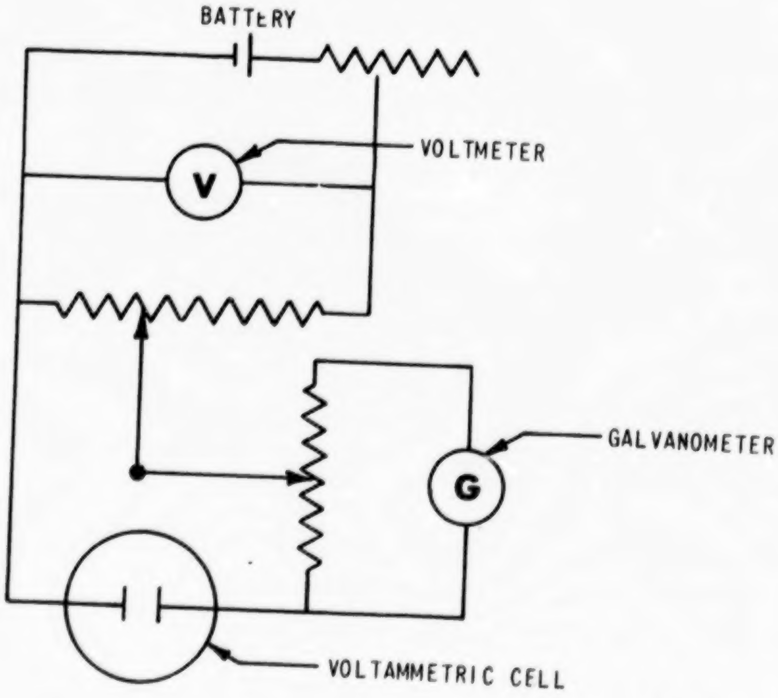
$$O_2 + 2H_2O + 2e \longrightarrow H_2O_2 + 2\,OH^- \qquad (36)$$

$$O_2 + 2H^+ + 2e \longrightarrow H_2O_2 \qquad (37)$$

$$O_2 + 4H^+ + 4e \longrightarrow 2H_2O \qquad (38)$$

A standard voltammetric system consists of a large nonpolarized reference electrode and a polarizable microelectrode (e.g., dropping-mercury electrode).

A variable potential is applied to the electrode system together with a sensitive means of measuring the current, as illustrated in Figure 21.



BATTERY
V
VOLTMETER
G
GALVANOMETER
VOLTAMMETRIC CELL

Figure 21. Voltammetric circuit.

The voltammetric current at constant voltage depends on the kinetics of the electrochemical reaction and on the rate of transport to the electrode of the substance involved in the reaction. Because the electrode reaction is usually rapid, at steady-state conditions the concentration of the electro-active species at the electrode surface is equal to zero. Accordingly, the rate-determining step in such electrode systems is that of mass transfer.

Mass transfer can occur as diffusion, convection, or migration. If electrolysis is conducted in the presence of a large excess of supporting electrolyte carrying virtually all the current, the migration effect can be eliminated.

Voltammetric electrode systems can be classified according to the mass transfer mechanism as:

1. Hydrodynamic and diffusion transport systems, e.g., rotating platinum electrode.

2. Infinite diffusion transport systems, e.g., dropping-mercury electrode (polarography).

3. Finite diffusion transport systems, e.g., membrane electrode.

Rotating Platinum Electrode

Electrode systems that make use of hydrodynamic and diffusion transport are composed of an electrode rotating in the test solution, or a stationary electrode in a stirred solution.

Rotating platinum electrodes have been used for several years for DO determination. The electrode system is composed of a platinum wire protruding a few millimeters from the wall of a glass tube, which rotates at a constant speed (e.g., 600 rpm). Electrical contact is obtained by filling the bottom of the tube with mercury and immersing another platinum wire (Figure 22).

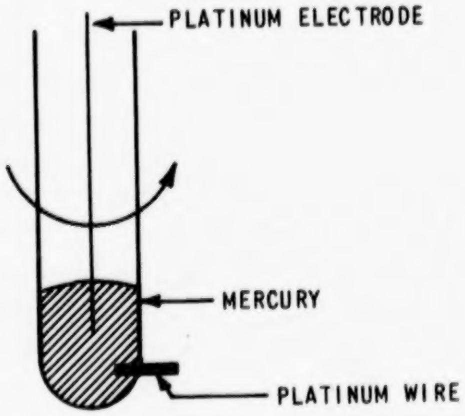


Figure 22. Rotating platinum electrode.

A typical current-potential curve for oxygen measured with a rotating platinum electrode is shown in Figure 23. The diffusion current, i_d, is directly proportional to the DO content. Reproducible current-potential curves are obtained provided the electrodes are kept clean and the same cleaning procedure is followed in comparative measurements.

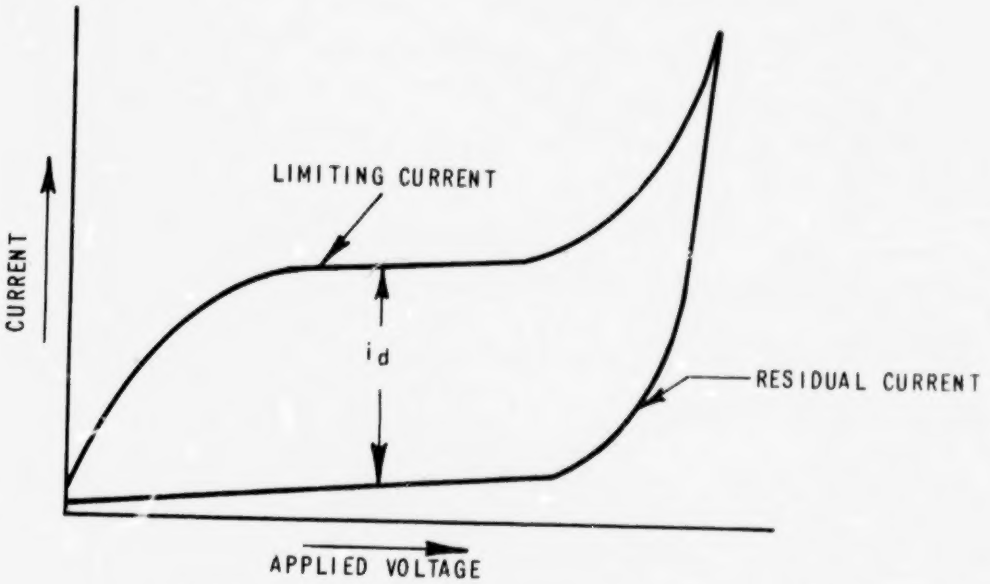


Figure 23. Oxygen polarogram.

Other systems employing hydrodynamic and diffusion transport are the vibrating platinum,[38] the spiral platinum,[22] the spherical platinum,[110] and the rotating-disc[14] electrodes.

Discussion - The use of the rotating platinum electrode offers the following advantages:

1. DO analysis can be performed *in situ*.
2. It is usable in continuous monitoring of "clean" waters.

The determination of DO by means of the rotating platinum electrode in many waters and wastes has not been feasible[66] because:

1. As the metal electrode comes in direct contact with the test solution, electroactive as well as surface-active compounds normally present in many waters and wastes may interfere with the electrode reaction. This is termed "electrode poisoning."
2. Occasional difficulty in maintaining a constant speed of rotation of the platinum electrode results in a distortion of the diffusion current.

Polarography

Polarography is a specialized voltammetric method, which makes use of diffusion transport in an infinite medium. The polarographic technique involving a dropping-mercury electrode has been in use for many years for the determination of DO in aqueous solutions. The technique is described in the 11th edition of Standard Methods[106] as a tentative procedure to be used when the Winkler Method and its modifications are subject to serious errors (e.g., in the presence of high concentrations of industrial wastes).

A polarographic assembly consists of a cathode in the form of a capillary tube attached to a vertical glass tube full of mercury and a mercury pool or standard calomel electrode as the anode. (Figure 24). The height of the mercury in the cathode tube is regulated to give the desired rate of flow. Short drop-time values, e.g., 3 to 4 seconds, are favored for DO determinations. The mercury emerging from the capillary tube in a steady flow of small drops forms a constantly renewed cathode surface.

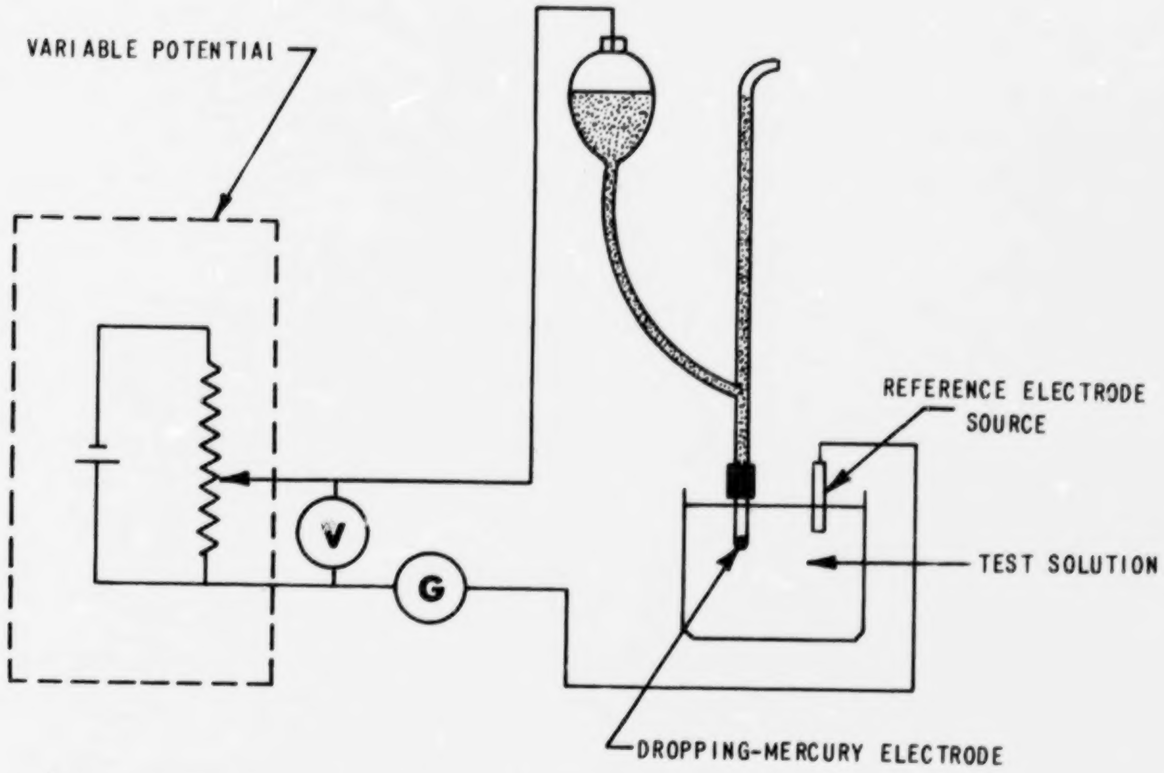


Figure 24. Polarographic circuit.

A typical oxygen polarogram (current-potential curve) is illustrated in Figure 25. The curve is composed of two waves representing the two steps of oxygen reduction at the dropping-mercury electrode. The first step of the electrolytic oxygen reduction, which has a half-wave potential of -0.1 volt versus the saturated calomel electrode (SCE) and a plateau in the potential range -0.3 to -0.6 volt versus SCE, conforms with the following reactions:

$$O_2 + 2H^+ + 2e \longrightarrow H_2O_2 \text{ (acid medium)} \quad (39)$$

$$O_2 + 2H_2O + 2e \longrightarrow H_2O_2 + 2OH^- \text{ (neutral or alkaline medium)} \quad (40)$$

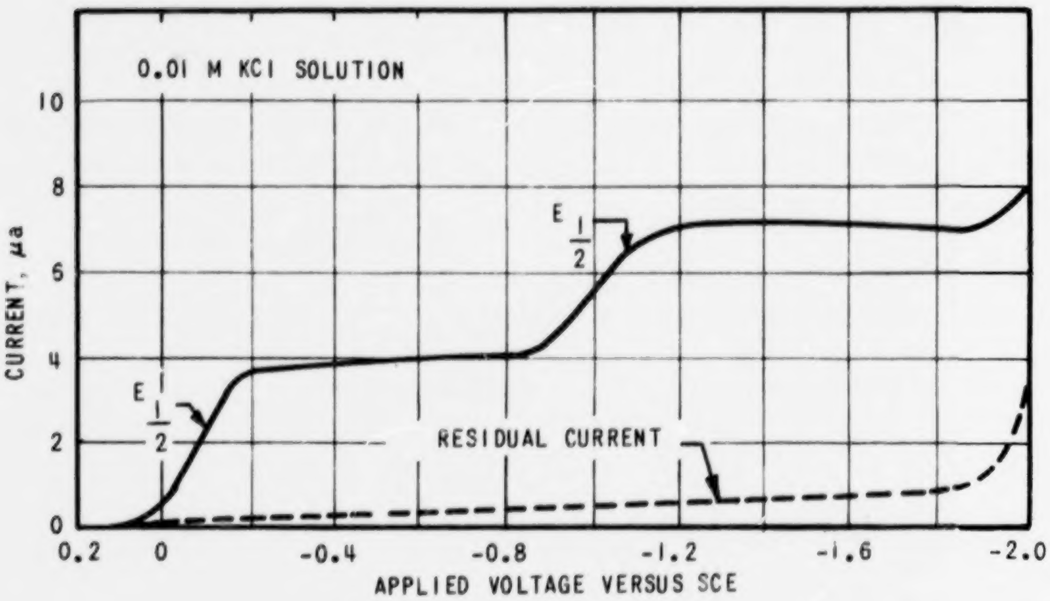


Figure 25. Oxygen polarogram.

The second step of oxygen reduction occurs at a more negative voltage, with a half-wave potential of -1.0 volt and a plateau at -1.6 volts versus SCE, and conforms to the following reactions:

$$H_2O_2 + 2H^+ + 2e \longrightarrow 2H_2O \text{ (acid medium)} \qquad (41)$$

$$H_2O_2 + 2e \longrightarrow 2OH^- \text{ (alkaline or neutral medium)} \qquad (42)$$

The diffusion current, i_d, the difference between the limiting current and the residual current, is directly proportional to the oxygen content in the test solution, C, as shown by the following equation:

$$i = 607\ n\ m^{2/3}\ t^{1/6}\ D^{1/2}\ C \qquad (43)$$

where: n = Number of electrons involved in the electrode reaction per mole of oxygen

m = Rate of flow of mercury, mg/sec

t = Drop time, sec

D = Diffusion coefficient for molecular oxygen, cm^2/sec

C = DO concentration, mM/liter.

For a given electrode system, n, m, d, and t are constant and equation (43) can be simplified to

$$i_d = \phi\ C_s \qquad (44)$$

where: ϕ = Sensitivity coefficient, μa/mM-liter.

The calibration of the instrument is preferably done with the solution to be tested. A plot of the diffusion current, at a certain applied voltage versus the DO concentration, determined by the Winkler Method, is shown in Figure 26.

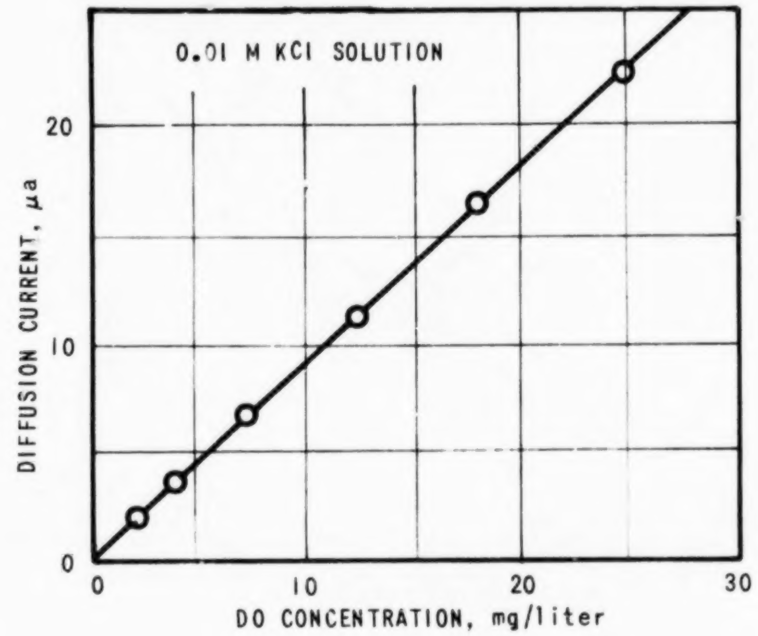


Figure 26. Calibration curve.

The choice of the operating voltage at which diffusion current measurements are taken is very important. It is desirable to use an applied voltage at which electroactive as well as surface-active contaminants do not interfere with the electrode reaction. This can be done by running an oxygen polarogram on a sample from the water to be tested. The shape of the curve makes it possible to find out the effect of contaminants and to choose an optimum operating voltage.[68]

The effect of surface-active agents[68] is illustrated in Figures 27 and 28. From Figure 27 it is clear that anionic surface agents (Aerosol O. T.) interfere with the electrode reaction in the potential regions corresponding to the first wave. The effect of cationic surface-active agents (e.g., Roccal, not usually found in natural waters) on the electrode reaction occurs in the potential region corresponding to the second wave as shown in Figure 28.

Standard Methods[106] recommends the use of an applied voltage corresponding to the first oxygen wave. This necessitates the addition of a maxima suppressor (surface-active agent) to the test solution. If, however, an applied voltage corresponding to the second oxygen wave is used (-1.6 vs SCE), a maxima suppressor will not be needed.[68]

In addition, anionic surface-active agents, e.g., alkyl benzene sulfonate (ABS), which interfere with the electrode reaction in a potential range corresponding to the first wave, do not have an effect at the second

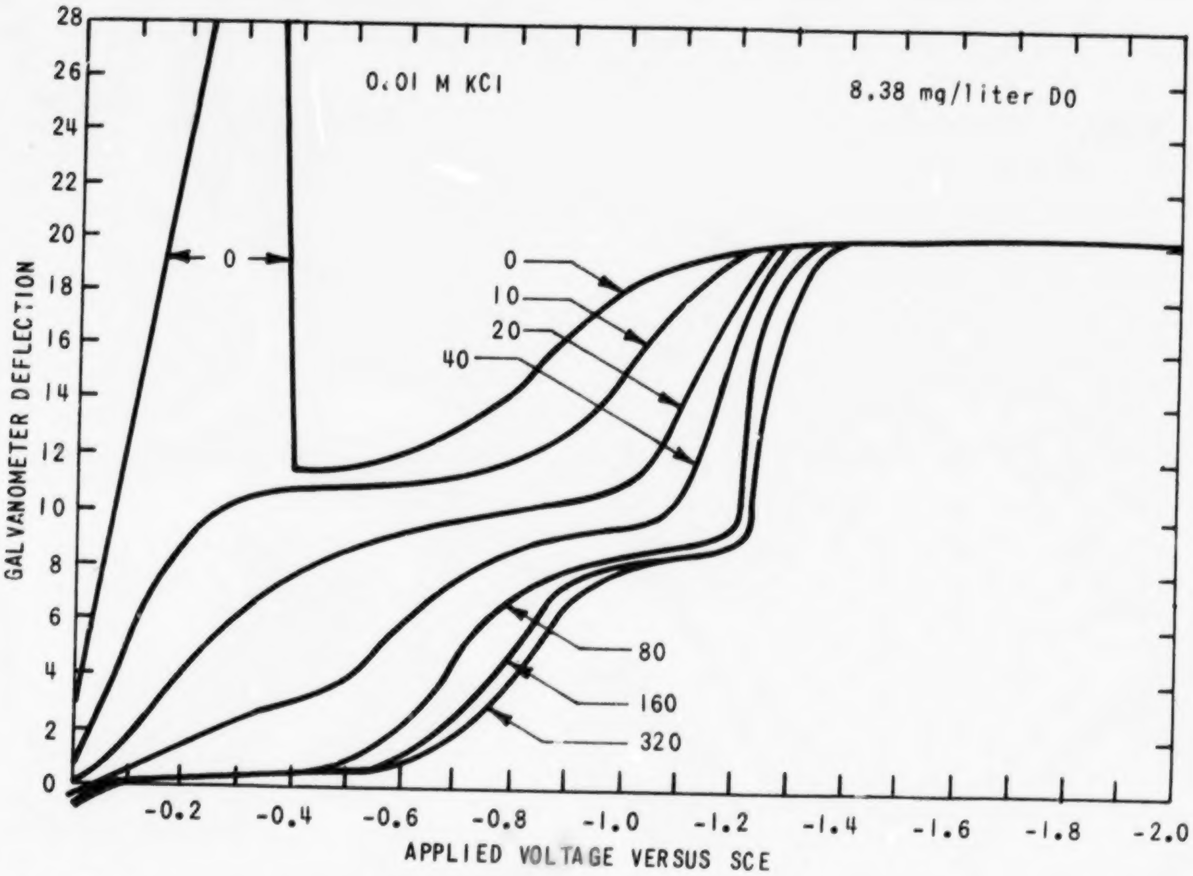


Figure 27. Oxygen polarograms at different aerosol O.T. concentrations (mg/liter).

oxygen wave.[68] A third advantage in using the second oxygen wave potential is that the oxidation waves of cyanides and sulfides will not interfere. The only source of interference that might be encountered at such negative potentials is the reduction of metal ions, such as zinc and nickel.

Several attempts have been made to adapt the dropping-mercury electrode for DO measurements in natural waters and waste waters.* In spite of the fact that DO can be measured accurately by conventional polarography, this method is not always reliable for continuous operation, especially in heavily polluted waters.[18, 93] Even in tap water, it was reported that the electrode system failed to function properly after a few hours of continuous operation.[18] This was explained by the deposition of calcium carbonate in or near the very fine bore of the capillary electrode. The deposits resulted in a change in the drop time and finally blocked the capillary completely. Frequent cleaning of the capillary tube should eliminate this difficulty.[11, 101]

In analysis of sewage and waste water, polarographic methods have not been reliable whenever continuous monitoring has been attempted.[4, 68] This deficiency is blamed on the presence of interfering materials, which are surface active or electroactive in nature, being adsorbed on or reacting with the mercury electrode. Recent studies[106] showed that the

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*See references 4, 17, 18, 19, 31, 46, 51, 53, 61, 73, 78, 84, 93, 101, 104, 119, 120.

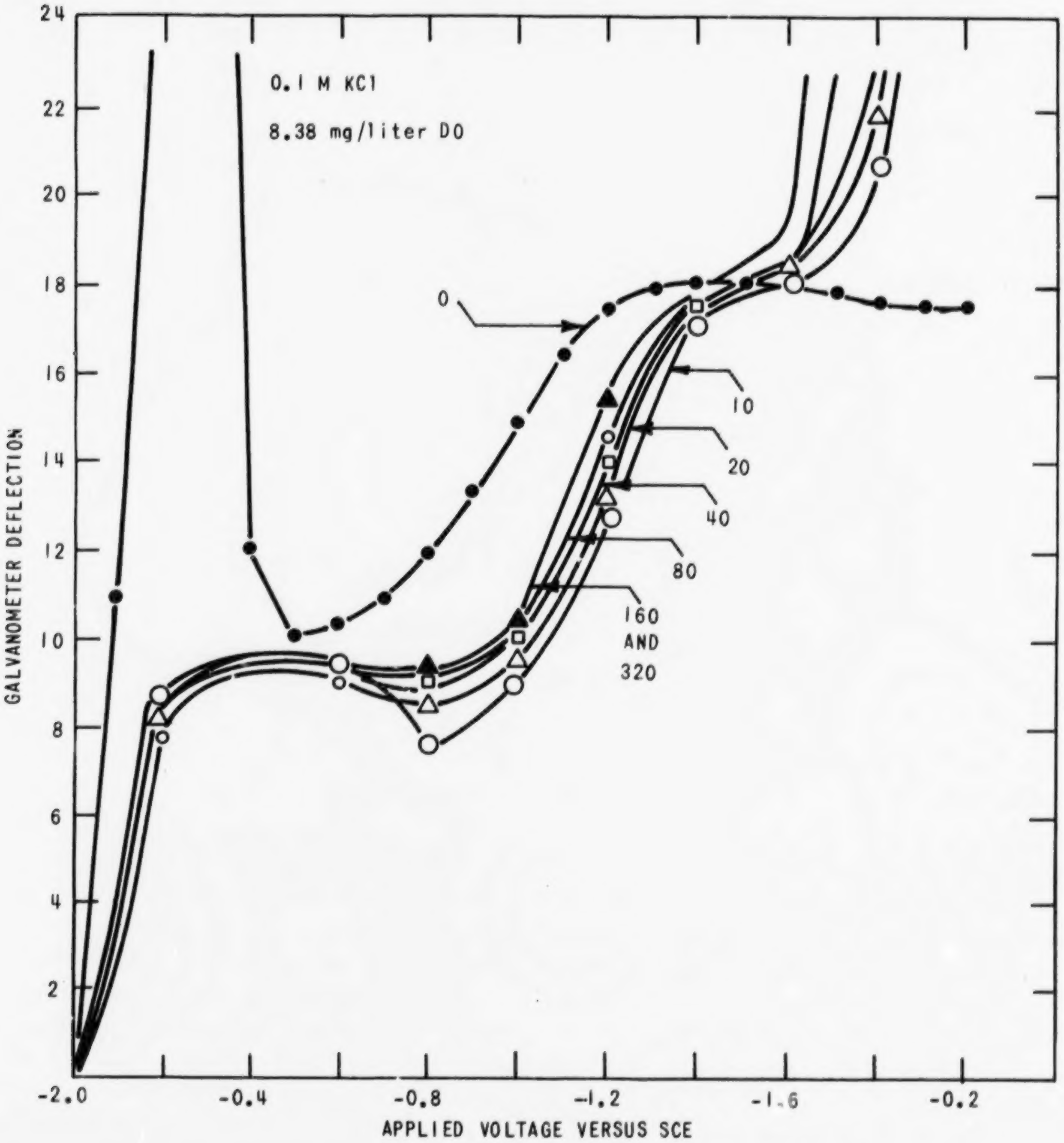


Figure 28. Oxygen polarograms at different Roccal concentrations (mg/liter).

adsorption of surface-active agents on the dropping-mercury electrode interfered with the electrode reaction only at the latter part of the drop-time curve, as shown in Figure 29. This figure indicates a time lag after which concentrations of surface-active agents above 20 mg/liter suppress the current. This time lag is needed to establish the adsorption equilibrium; its magnitude depends on the concentration and the type of surface-active agent used. It is obvious then that the shorter the drop time, the less the effect of surface-active agents on the electrode reaction. Rapid-dropping-mercury electrodes,[52, 120] sometimes called wide-bore dropping-mercury electrodes, have been used with considerable success. This new type of dropping-mercury electrode consists of an upward-sloping capillary of 0.8-mm internal diameter, and a zinc anode,[13] as shown in Figure 30. The mercury flow rate, m, in such

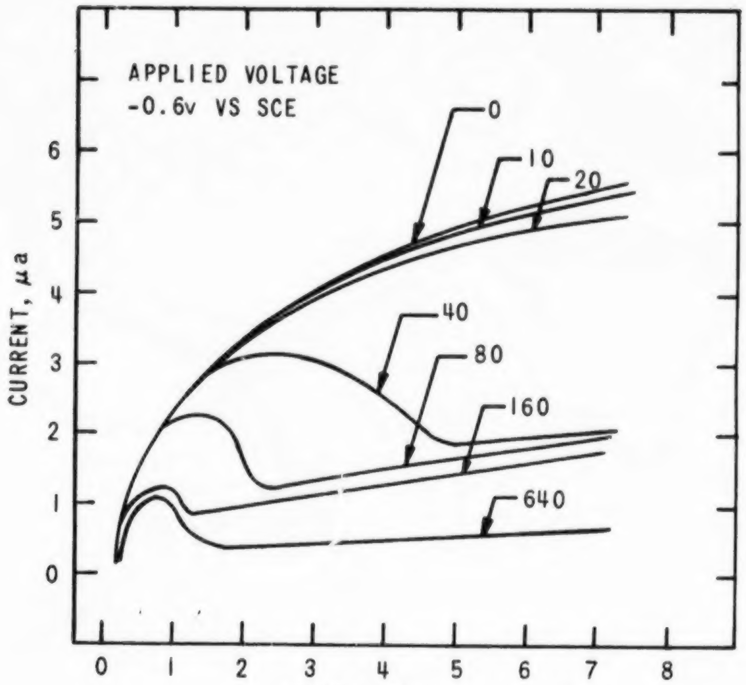


Figure 29. Current-time curves at different ABS concentrations (mg/liter).

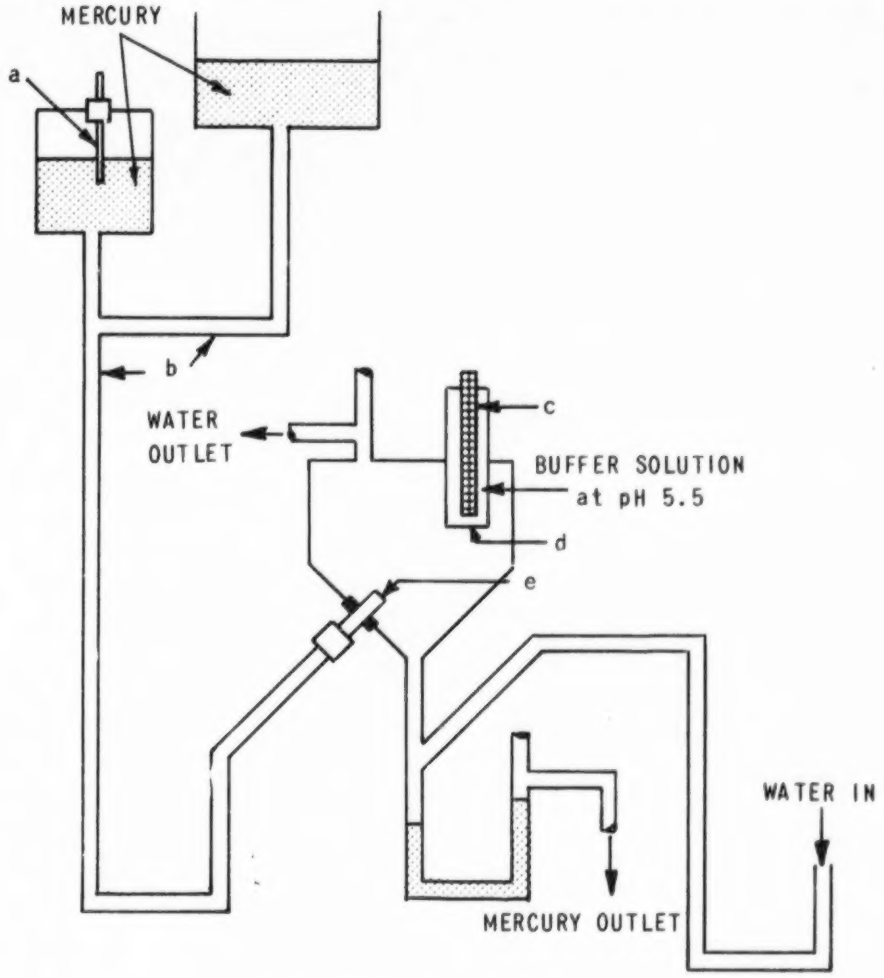
electrode systems is about 7 mg/sec, with a drop time, t, of 0.20 to 0.33 sec.[119]

Discussion -- The use of rapid-dropping-mercury electrodes for DO measurements provides the following advantages:

1. The use of the short drop time minimizes the effect of surface-active agents on the electrode reaction.
2. The rapid-dropping-mercury electrode can be used in flowing solutions or with stationary water samples.
3. This method results in higher sensitivity because the current is about 10 times greater than with the conventional electrode system.

Drawbacks of polarography for the determination of DO in natural waters and wastes are:

1. Although dropping-mercury electrodes have an advantage of a continuously renewed surface, the electrode system lacks the ruggedness and portability required for a field instrument.
2. In the polarographic technique the sensing electrode comes in direct contact with the test solution. As a result certain electroactive as well as surface-active species in the test solution may react with or adsorb on the electrode surface and cause serious interferences with the DO measurement.
3. The toxic effect of mercury on biological systems makes this technique unsuitable for the determination of oxygen respiration or oxy-



a = PLATINUM CONTACT
b = SILICON RUBBER TUBING
c = ZINC ELECTRODE
d = No. 4 SINTERED GLASS DISC
e = DROPPING-Hg ELECTRODE

Figure 30. Rapid-dropping-mercury electrode.

gen uptake studies.

The accuracy of the method[106] depends to a great extent on the procedure used. In the absence of interferences, the accuracy ranges between 0.05 to 0.10 mg/liter with a precision of ± 0.01 mg/liter.

Galvanic Methods

Oxygen-sensitive galvanic cells have been known for a long time. Air cells, cells utilizing atmospheric oxygen, were used as sources of electrical energy prior to the invention of the Leclanche dry battery.

The classic air cell is composed of a zinc anode or anodes and a porous carbon cathode immersed in 20 percent aqueous caustic soda. In contrast to the Leclanche dry cell, the electrolyte in the air cell takes a prominent part in the cell reaction, and the life of the cell finally ceases because of exhaustion of electrolyte.

If, however, a specially designed air cell is used, the generated current will be proportional to the oxygen concentration in contact with the cathode.[5, 6, 109, 115-117, 126] A variety of galvanic couples and modifications of the electrode system has been reported. A typical analyzer in which the oxygen-sensing element is a galvanic cell is shown in Figure 31.

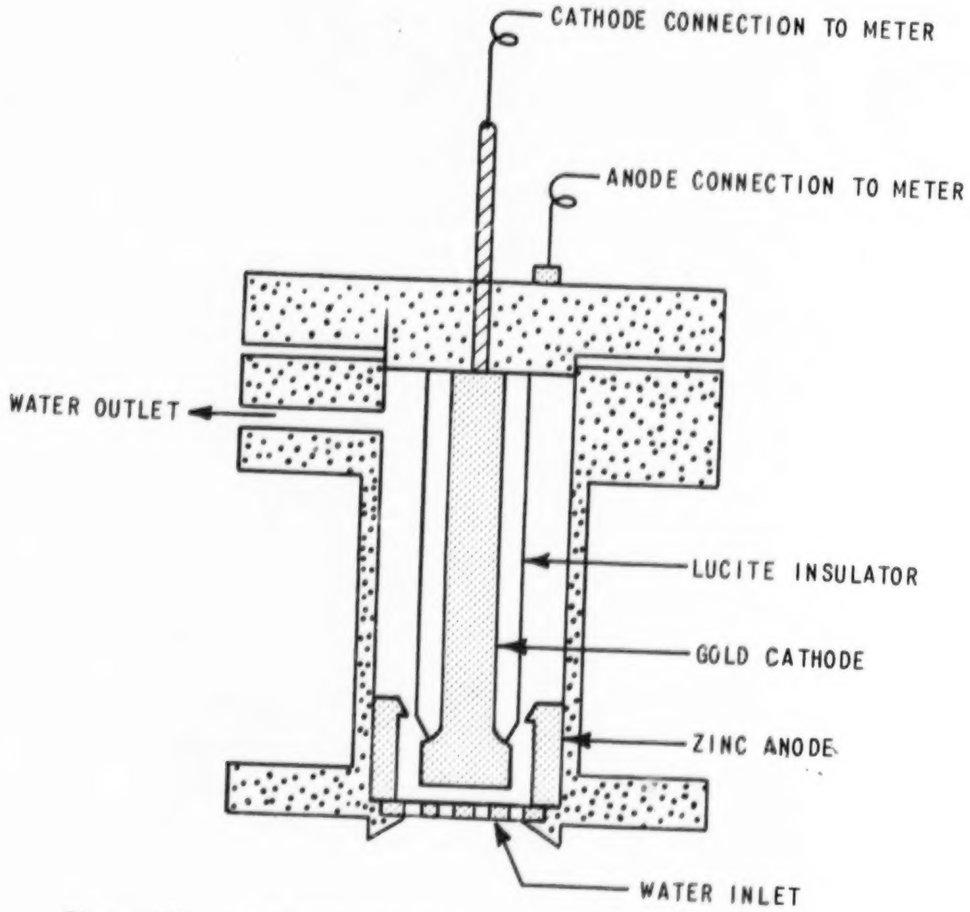


Figure 31. Hartmann and Braun dissolved oxygen analyzer.

Changes in the pH and the conductivity of the test solution influence the DO measurements. Instruments with appropriate circuits adjustable for conductivity were recently developed.[115] For water samples with very low conductivity, gaseous compounds are added to increase the conductivity, e.g., ammonia, gaseous amines, or carbon dioxide.[117]

Since the early 1930's[116] galvanic methods for the analysis for DO have been used. Several varieties of galvanic systems utilizing carbon, gold, or platinum cathodes are used in Europe.[116] The usefulness of this type of cell is limited because the electrode system may be easily poisoned. This is evident since the electrode system comes in direct contact with the test solution and surface-active compounds as well as other suspended material frequently adsorb on the electrode surface. This is particularly true in waste waters. To prevent an incrustation, a small amount of HCl may be added to the water by means of a dosing device ahead of the galvanic cell.[116]

DO in activated-sludge units has been determined by a specially designed galvanic system.[109] The instrument used (Figure 31) consists of a circular gold (or platinum) cathode of limited area and a large zinc anode dipped into a swift-flowing stream (1 m/sec) of the liquid to be tested. To keep the electrode surfaces clean, they are rotated against a scraper.

Discussion - Galvanic methods of analysis offer the following advantages:

1. Galvanic cells are capable of oxygen determination in situ.
2. They are capable of continuous monitoring.

The main disadvantages are:

1. Since the sensing element comes in direct contact with the test sample, one of the principal difficulties has been with impurities that either react with or adsorb on the electrode surface.
2. Changes in the conductivity of the water, such as in estuarine water, will cause a change in the sensitivity of the analyzer.

MEMBRANE ELECTRODES

Types of Membrane Electrodes

Perhaps the most dramatic development in the analysis for DO since the Winkler Test is the utilization of membrane electrode systems. These are particularly suitable for applications in environmental science and engineering. Only within the last 3 to 5 years, however, have membrane electrode systems been used for DO determinations in natural and waste waters.

Membrane electrode systems used in the analysis for DO can be described as finite-diffusion electrochemical cells.

To understand the principle of these electrochemical cells it may be helpful to consider conventional polarographic systems.

In polarography the diffusion current is solely dependent on the rate of diffusion of the electroactive species in the test solution. The test solution being of no definite volume, the transport phenomenon is that of infinite diffusion. In membrane electrode systems the diffusion current is solely dependent on the rate of diffusion of the electroactive species in a finite layer, which is achieved by placing a plastic membrane in close contact with the electrode surface so that it separates the electrode system from the test solution. Under these conditions the membrane constitutes a rigorously defined diffusion layer, the thickness of which is independent of the hydrodynamic properties of the test solution. Such electrode systems exhibit more stable performance characteristics than the conventional types.

Finite-diffusion membrane electrode systems are of two main types, voltammetric membrane electrodes and galvanic membrane electrodes. The basic difference between voltammetric and galvanic membrane electrode systems is that the former rely on an external source of applied voltage to polarize the indicator electrode, similar to the conventional polarographic analysis. Galvanic analysis[43] is defined, on the other hand, as a method that makes the species of interest a reactant at one of a pair of electrodes, thereby generating an electrical current without the aid of an electromotive force. Hence in the presence of the substance under galvanic analysis, a combination of electrodes (galvanic couple) and an electrolyte, the galvanic system becomes a primary battery or fuel cell in the general sense.

Several membrane electrode systems have been developed and used for the analysis for DO in natural waters.[23, 25]

Recently, a galvanic membrane electrode system[70] has been used widely and successfully in water pollution control programs and in waste treatment operations.[69, 72] This electrode system is shown in Figure 32. It is composed of a silver-lead galvanic couple arranged in a ring-shaped anode surrounding a central disc-shaped silver cathode.[71, 72] The galvanic couple is fitted to the tip of the plastic probe and is covered by a 0.5- or 1.0-mil-thick polyethylene membrane. A small amount of 4 M KOH, held by a small disc of lens paper, serves as a layer of electrolyte solution between the membrane and the electrode surface. After 2 to 4 weeks of operation the electrode system is recharged by cleaning the electrode surfaces, replenishing the supporting electrolyte, and mounting a new membrane.

With respect to circuitry, galvanic sensors are as simple to work with as thermocouples. All that is needed with the galvanic cell oxygen analyzer is a low-impedance galvanometer or microammeter. For recording, an inexpensive galvanometer-type recorder is adequate in most cases. Also, the cell circuit may be closed with a known resistor and the potential drop fed to a potentiometric recorder. Integrators, alarms, and controller equipment can be operated.

The load resistance in the circuit has little effect on the cell sensitivity since it is in series with the large polarization resistance of the sensing electrode. At high values of external load, however, the galvanic current is affected.

A galvanic membrane electrode that can be sterilized with steam has been used successfully for DO determinations in aerobic fermenters.[51] This electrode system is made of a silver-lead galvanic couple and an acetate buffer as the supporting electrolyte.

Galvanic membrane electrodes, besides being self-energizing systems, have the advantage of being self-zeroing. In voltammetric membrane electrodes, an impressed voltage of either sign may give background signals.[43]

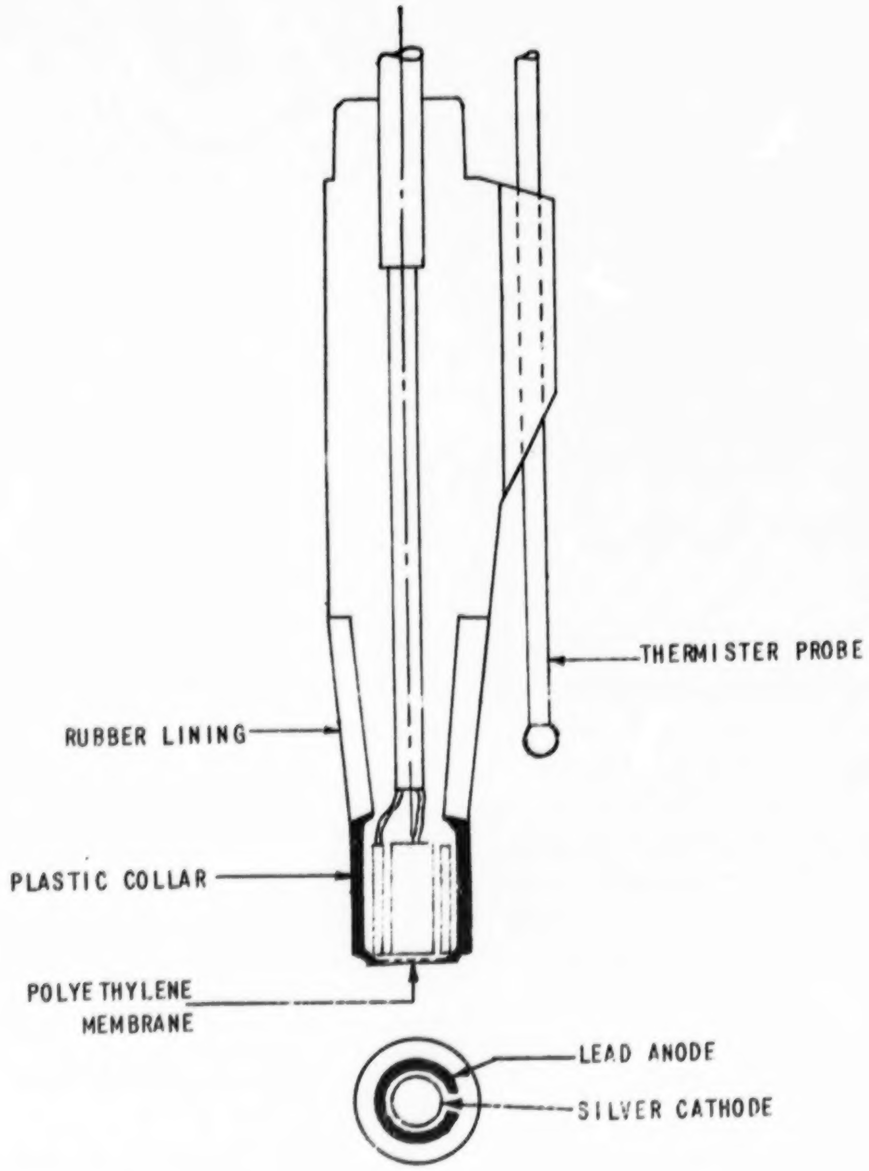


Figure 32. Galvanic cell oxygen analyzer.

Principle of Oxygen-Membrane Electrodes

There are three main steps involved in the operation of oxygen membrane electrodes. The first is the permeation of molecular oxygen from the test solution through the plastic membrane layer. The second is the permeation of molecular oxygen through the electrolyte solution layer. The third step is the electrolytic discharge of oxygen on the silver cathode, with the subsequent generation of an equivalent quantity of current.

To aid in understanding of the theory involved, the diffusion current equations as reported by the senior author[71] are presented briefly. Figure 33 shows a cross-sectional diagram normal to the surface of the indicator electrode, which is composed of an electrolyte solution layer of thickness "A" and a membrane layer of thickness "B." Concentration profiles at different times of electrolysis, t, are also shown in Figure 33.

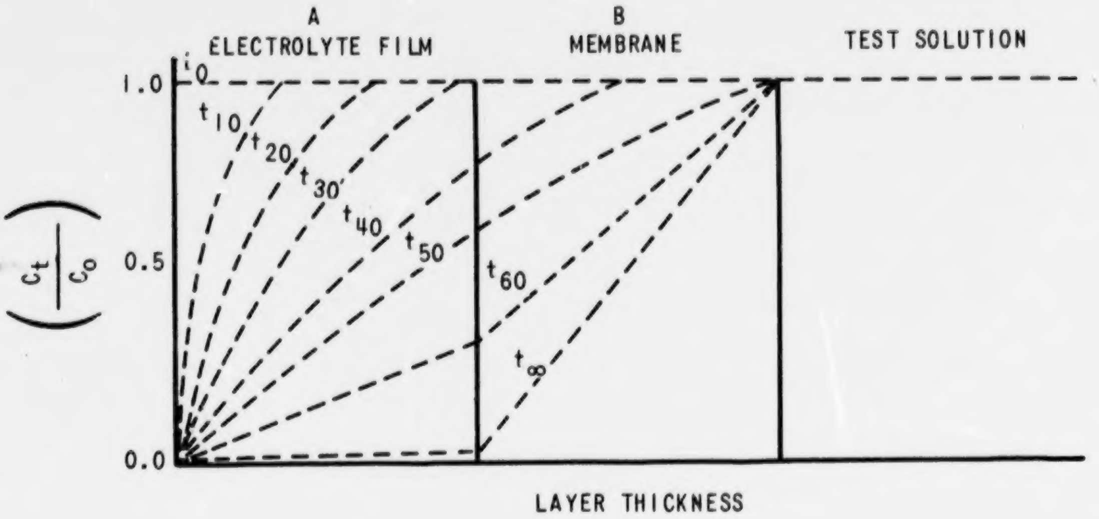


Figure 33. Concentration profiles.

Figure 34 shows the corresponding current-time curve, in which steady-state operation was reached after 90 seconds. The current is assumed to be solely dependent on the rate of mass transfer (linear finite diffusion) of reactants to the electrode surface and not on the overall charge-transfer rate or on the kinetics of a chemical reaction coupled with the charge-transfer process. As soon as the electrolysis circuit is closed, the current shoots up to a high initial value, which is characteristic of charging the capacitance of the double layer (Figure 34), after which the current decreases in a manner determined by the rate of mass transport, as illustrated by the concentration profiles in Figure 33. Current measurements at t = 10 seconds, corrected for capacitance current thus depends on the concentration of the electroactive species in the electrolyte solution layer only. This can be easily seen on considering the diffusion current equations for such two-film (the electrolyte film and the membrane) electrode systems.[71] At very small values of t (e.g., below 30 seconds in Figure 33), the electrolyte film is of primary importance and the current varies according to the following equation:

$$i_t = nF\bar{a}\left(\frac{D_f}{t}\right)^{0.5} C\left\{1 + 2\sum_{n=0}^{\infty} e^{-\frac{n^2A^2}{D_f t}}\right\} \qquad (45)$$

where: i_t = Current at time t, μa

n = Number of electrons exchanged per mole of reactant

F = Faraday = 96,500 coulombs

$\bar{a}$ = Surface area of indicator electrode, cm^2

D_f = Diffusion coefficient of reactant in electrolyte solution layer, cm^2/sec

t = Time, sec

C = Concentration (strictly the activity) of reactant species in test solution, M/cm^3

A = Thickness of electrolyte solution film, cm.

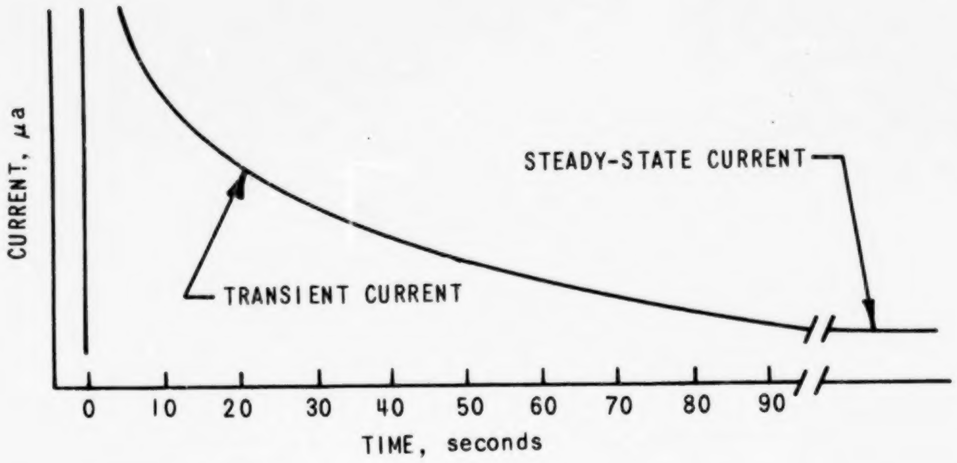


Figure 34. Current-time curve.

At very small t values, the summation term in equation (45) can be neglected and the current varies inversely with the square root of time.

$$i_t = \left[nF\bar{a} \left(\frac{D_f}{\pi} \right)^{0.5} C \right] t^{-0.5} \qquad (46)$$

At larger values of t ($t \geq 30$ sec in Figure 33), diffusion in the membrane is governing.

$$i_t = nF\bar{a} \frac{P_m}{B} C \left\{ 1 + 2 \sum_{n=0}^{\infty} e^{-\frac{n^2 \, {}^2 D_m t}{B^2}} \right\} \qquad (47)$$

where: P_m = Membrane permeability coefficient, cm^2/sec

D_m = Diffusion coefficient of reactant in film, cm^2/sec

B = Thickness of membrane layer, cm.

At large values of t (above 90 sec), steady-state conditions are achieved and equation (47) is then:

$$i_\infty = nF\bar{a} \frac{P_m}{B} C \qquad (48)$$

The rate determinant in this case is the transport through the membrane. Accordingly, the generated current is directly proportional to the rate of permeation of molecular oxygen through a given membrane, which, in turn, is directly proportional to the DO content of the test solution. A typical calibration curve showing the linearity of response under steady-state conditions is shown in Figure 35.

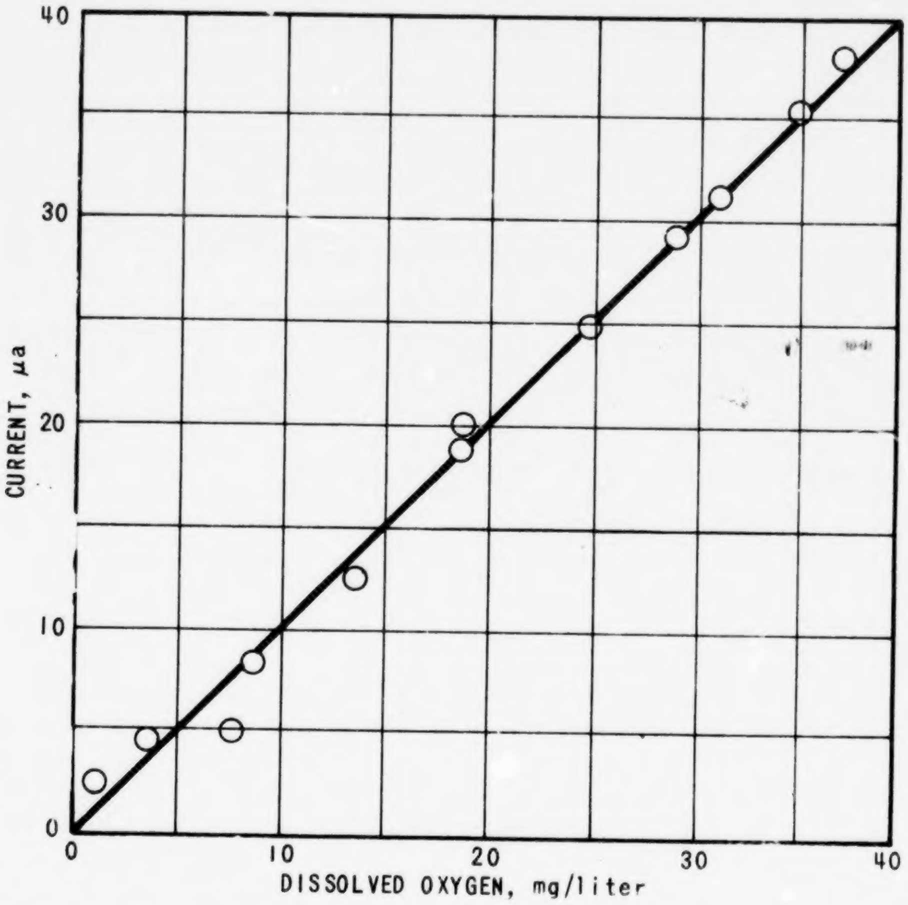


Figure 35. Calibration curve at 25°C.

Unpublished data from recent studies by the senior author revealed that after the first day of continuous operation, the electrode reactions of the galvanic cell oxygen analyzer are partially as follows:

Cathodic reaction -

$$\tfrac{1}{2}O_2 + H_2O + 2e \longrightarrow 2OH^- \qquad (49)$$

Anodic reaction -

$$Pb + 2OH^- \longrightarrow PbO + H_2O + 2e \qquad (50)$$

Overall reaction -

$$Pb + \tfrac{1}{2}O_2 \longrightarrow PbO \qquad (51)$$

It is evident then that as the cell discharges, there is no net consumption of the OH^- concentration. Although, the OH ions are not consumed in the overall cell reaction, they are required to sustain the anodic reaction. Hence there will be a localized defficiency of OH^- ions at the anode and an increase in the OH^- ions in the electrolyte solution layer from the central cathodic area toward the peripheral anode.

Oxygen membrane electrodes have three main components: the membrane, the oxygen-sensing element, and the electrolyte solution. The membrane, the unique feature in such electrode systems, serves in three different capacities. First, the membrane acts as a protective diffusion barrier separating the sensing element from the test solution. Since plastic membranes are permeable to gases only, oxygen molecules pass through, but electroactive and surface-active contaminants present do not. The possibility of poisoning the sensing element is thus eliminated.

Second, the membrane serves to hold the supporting electrolyte in contact with the electrode system and thus makes it possible to determine oxygen in gaseous samples as well as in nonaqueous solutions such as industrial wastes.[71]

The third advantage, discussed previously, is that the membrane constitutes a finite diffusion layer, the thickness of which is independent of the hydrodynamic properties of the test solution.

Membranes must meet the following requirements. Primarily, the membrane should have excellent permeability to oxygen yet be practically impermeable to ionic species and water molecules from the test solution. Its permeability characteristics should not change with time, and there should be no interaction between the membrane material and the diffusion oxygen molecules or ionic species present in the test solution. The membrane should be able to withstand slight amounts of stretching without breaking so that it can be applied easily to the electrode surface.

Polyethylene, Teflon, polypropylene, and synthetic rubber show good permeability for oxygen. Polyethylene membranes of low crystallinity were found to have high permeability to oxygen.[72]

As far as the oxygen-sensing element is concerned, platinum has been used extensively as cathode material in electrochemical analysis for DO. Recent investigations[71] reveal that silver and gold are better cathode materials for oxygen detection than the conventionally used platinum. The reduction of oxygen on a platinum cathode was found to be a complicated process since the reaction proceeds through the formation of a platinum oxide layer.[62] Significantly, this step does not occur with silver or gold electrodes.

The electrolyte solution in membrane electrode systems should be a highly conducting aqueous solution that does not interact with the electrode material. In voltammetric systems the type and concentration of the electrolyte solution are dictated by the reference anode, e.g., for an Ag/AgCl reference electrode, a 3 percent KCl solution is used.

Performance Characteristics

Sensitivity - The membrane oxygen electrode is commonly calibrated by means of the Winkler Test. A typical calibration curve is given in Figure 35, which shows a linear response to oxygen from zero to saturation values.

A basic operational requirement for membrane electrode systems is the maintenance of a threshold amount of mixing in the test solution around the tip of the analyzer. This can be achieved either by driving the solution past the surface of the analyzer by means of a pump or a stirring device, or by moving the analyzer. In the laboratory, a magnetic stirrer or a motor-driven glass stirrer is usually used. Since the flow past the probe must be not less than 1 ft/sec, appropriate flow in the field may be attained by the natural flow of the water, by towing the probe behind a moving boat, or by moving the probe up and down manually. Attachments containing battery-driven small propellers[49] may be mounted on the tip of the probe.

The effect of stirring the test sample on instrument sensitivity is shown in Figure 36. From an initial low value with no stirring, the sensitivity rises rapidly with increasing stirring speed and then assumes a steady value. Adequate stirring of the test solution is necessary so that the oxygen concentration at the membrane surface will be equal to that in the bulk of the solution. Only under such conditions will the measured diffusion current be directly proportional to the oxygen concentration in the bulk of the test sample.

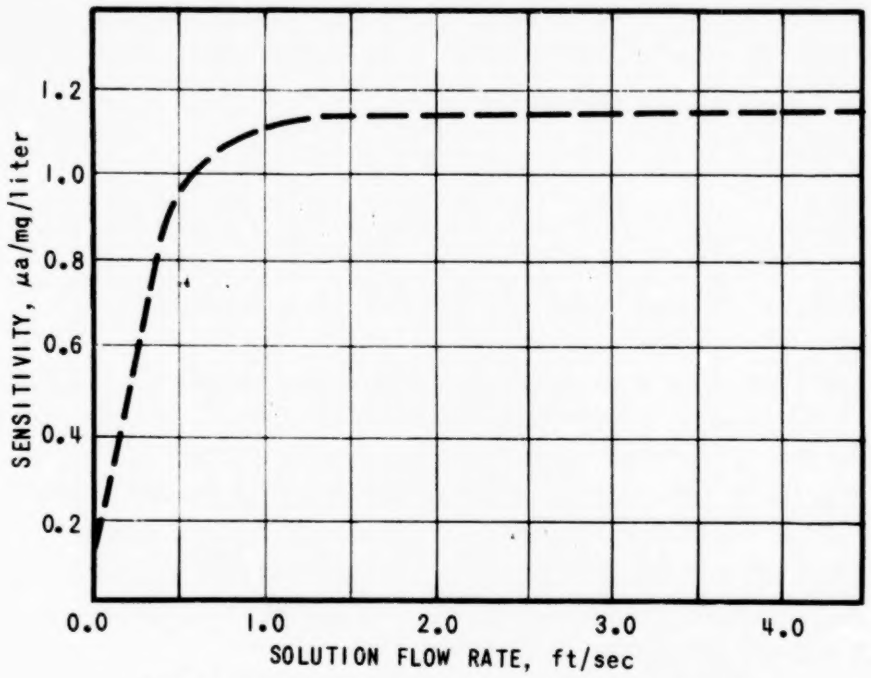


Figure 36. Effect of stirring on sensitivity.

Temperature Coefficient - Another critical characteristic of voltammetric membrane electrodes is the relatively high temperature coefficient, which is largely due to the effect of temperature changes on the permeability characteristics of the plastic membrane. Temperature effects on the electrode reaction kinetics or the electrode potential are relatively negligible. Calibration curves for one of the galvanic cell oxygen analyzers over the temperature range from 5° to 35°C are shown in Figure 37.

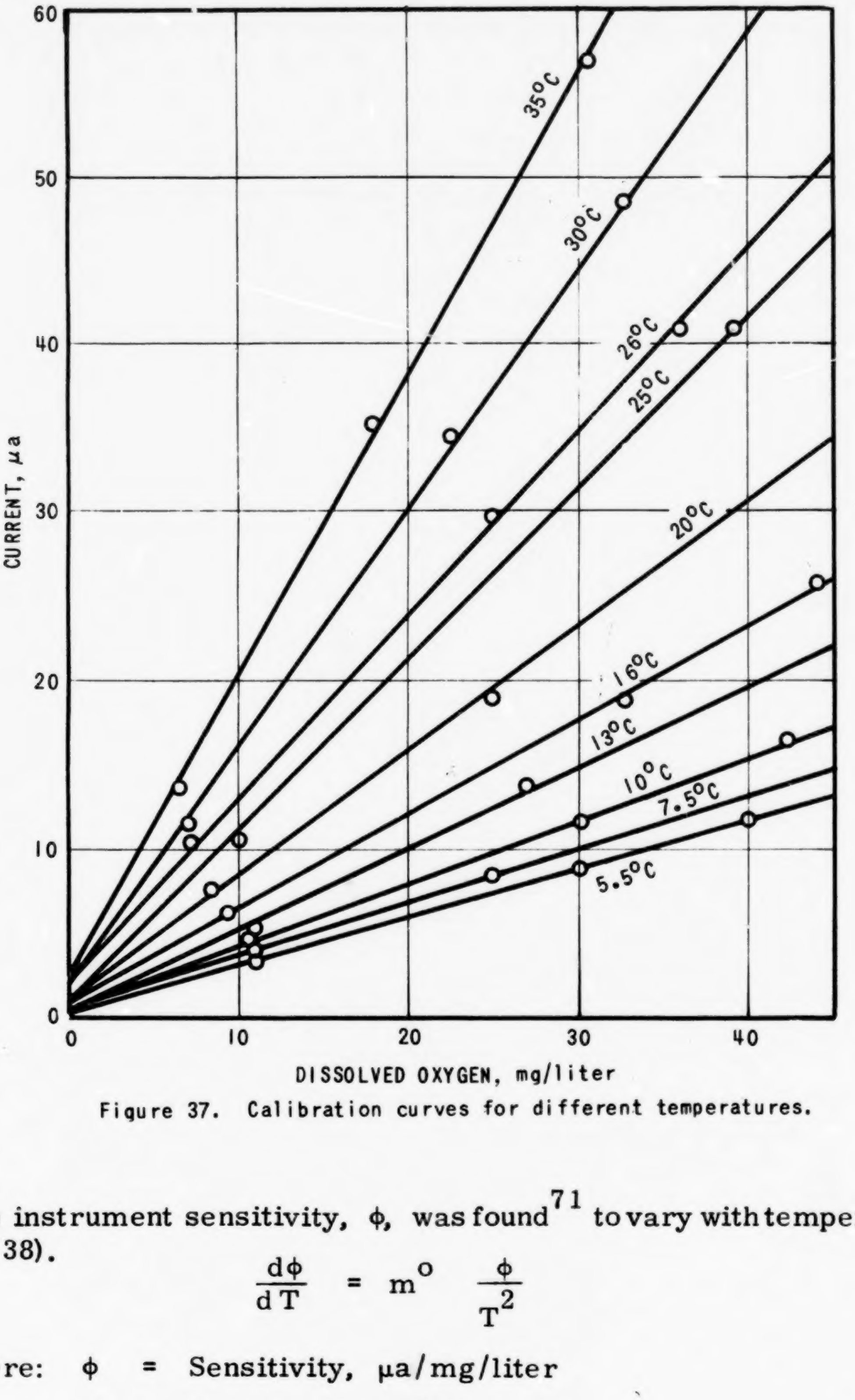


Figure 37. Calibration curves for different temperatures.

The instrument sensitivity, φ, was found[71] to vary with temperature (Figure 38).

$$\frac{d\phi}{dT} = m^{o} \frac{\phi}{T^{2}} \qquad (52)$$

where: ϕ = Sensitivity, μa/mg/liter

T = Temperature, °K

m^{o} = Temperature coefficient, °K

If log ϕ is plotted versus $1/T$, a straight line is obtained.

$$\log \phi = \frac{-m^o}{2.303} \frac{1}{T} + b \tag{53}$$

in which $-m^o/2.303$ is the slope and b is the ordinate intercept. The theoretical significance of this equation is discussed elsewhere.[71, 72] If m^o is determined, the instrument sensitivity can be calculated for any desired temperature.

$$\log \phi = \log \phi_o + \frac{m^o}{2.303} \left(\frac{1}{T_o} - \frac{1}{T}\right) \tag{54}$$

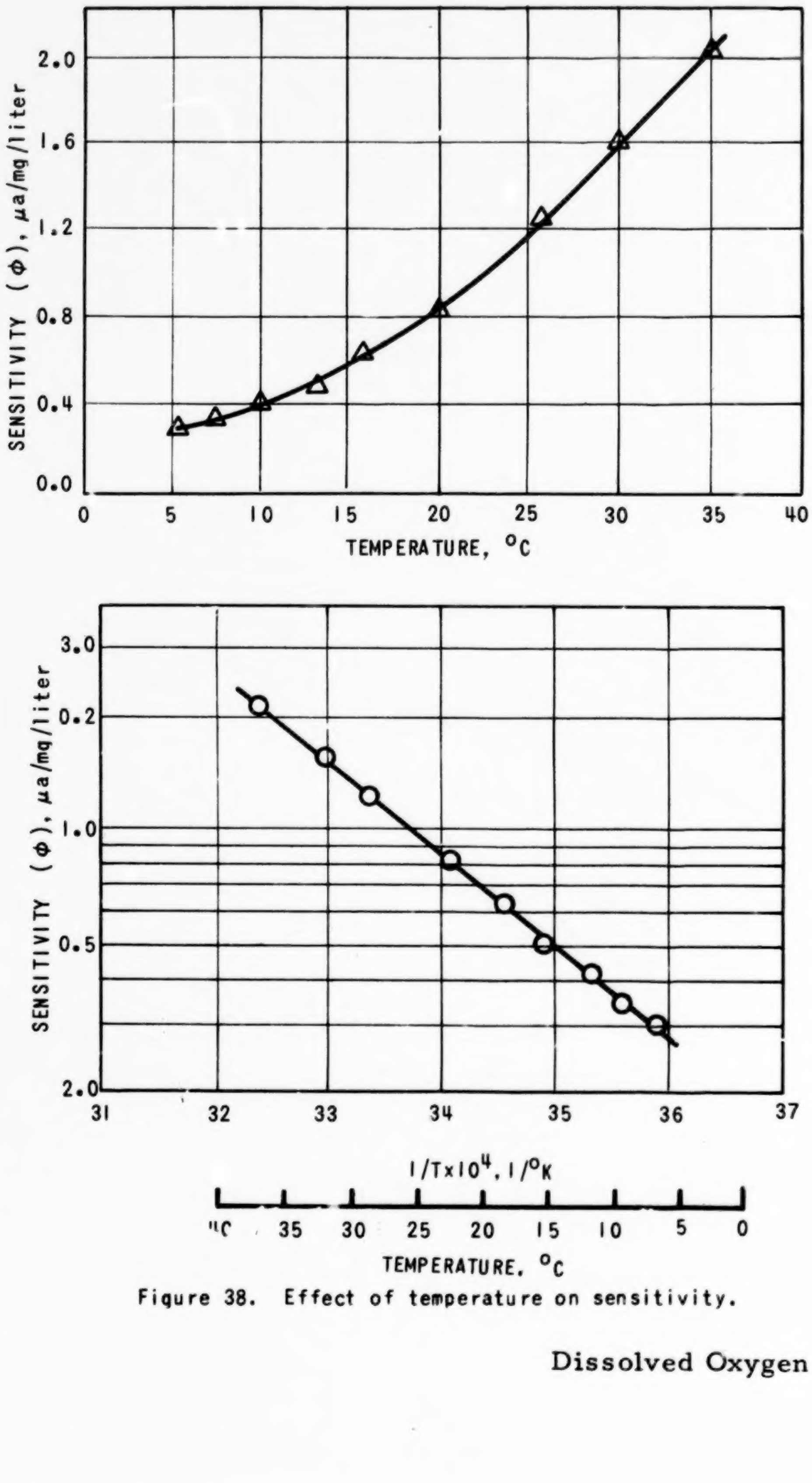


Figure 38. Effect of temperature on sensitivity.

Nomograph charts also can be constructed for the temperature correction.

Temperature compensation can be done automatically by incorporating a thermistor setting in the electrode circuit.[23] Thermistors do not compensate fully for the temperature errors to less than ± 10 percent of the reading over the temperature range of 5° to 35°C, or less than ± 5 percent over the 15° to 45°C range.[16] When high accuracy is required, calibrated nomographic charts should be used. Electrode systems with attached thermistors are used to obtain simultaneous temperature readings.[37]

Discussion

Interpretation of the results of oxygen membrane electrode systems requires careful consideration. It is helpful to point out the similarity between the glass membrane electrodes, used for the measurement of pH and pNa, and oxygen membrane electrodes. The measured parameter in the glass electrode system is an asymmetry potential across the glass membrane. This potential across the glass membrane is related to the hydrogen ion activity in the test solution.

In the case of oxygen membrane electrode systems, the measured diffusion current is solely dependent on the difference in the chemical potential of the electroactive species across the membrane. One should expect then that as glass electrodes are used for pH determinations (intensity), oxygen membrane electrode systems will likewise measure an "intensity property" that is essentially equivalent to the activity of molecular oxygen in solution. This should be differentiated from "extensive parameters" as determined by titrimetric methods such as the Winkler Test. Accordingly, the difference between the pH value determined by the glass electrode and acidity as determined by titration with a standard base is essentially the same as the difference between oxygen analysis by a membrane electrode and the Winkler Test. Only under ideal conditions and in the absence of certain impurities are diffusion current values from membrane electrode systems proportional to concentration. The correct form of the diffusion current equation for membrane electrodes is thus:

$$i = \left(nF\bar{a}\ \frac{P_m}{B}\right) a = \phi_a\ a = \phi_a\ \gamma C \qquad (55)$$

where: ϕ_a = Sensitivity coefficient with respect to activity, $\mu a/M/cm^3$

γ = Activity coefficient of molecular oxygen

n = Number of electrons exchanged per mole of reactant

F = Faraday = 96,500 coulombs

$\bar{a}$ = Surface area of indicator electrode, cm^2

B = Thickness of the membrane layer, cm

P_m = Membrane permeability coefficient, cm^2/sec

a = Activity of molecular oxygen, M/cm^3

C = Concentration of molecular oxygen, M/cm^3.

The dependence of the diffusion current on the ionic strength[71] of the test solution (Figure 39) can be expressed as follows:

$$i = (e^{K_s I} \phi_a)\ C \qquad (56)$$

where: K_s = Salting-out coefficient

I = Ionic strength.

Equation (56) correlates the diffusion current, i, to the concentration of molecular oxygen in the test solution as a function of the ionic strength, I. In the case that I = 0, e.g., distilled water, then the term $e^{K_s I}$ will be unity and equation (56) will be equal to equation (55). By means of equations (55) and (56) values can be determined for the activity coefficient, γ, and the salting-out coefficient, K_s, in salt solutions (e.g., KCl, $CaCl_2$, Na_2SO_4), which are of high theoretical significance.

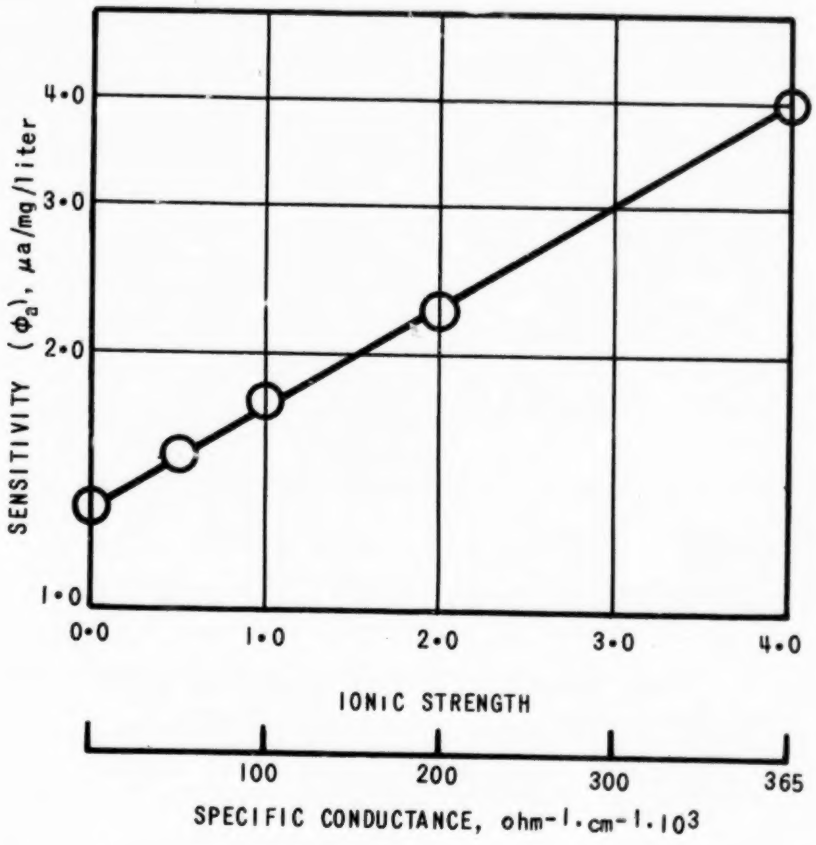


Figure 39. Effect of ionic strength on sensitivity.

Recent investigations have shown that simple methods can be developed to compensate for the temperature and the salt effects on the determination DO concentration by membrane electrodes.

The temperature effect[71] can be described as follows:

$$i = \phi_{o,t}\, e^{-\frac{E_p}{R} \Delta \frac{1}{T}}\, C \qquad (57)$$

where: $\phi_{o,t}$ = Sensitivity (current in amperes/DO concentration in M/liter) at a reference state with respect to temperature in °C

E_p = Activation energy of permeation of membrane, cal/M

T = Temperature, °K

R = Gas constant, cal/M/°K

C = Concentration of molecular oxygen, M/liter.

Similarly the effect of the salt content of the test solution can be expressed as follows:

$$i = \phi_{o,s}\, e^{K_s \Delta I}\, C \qquad (58)$$

where: $\phi_{o,s}$ = Sensitivity at a reference state with respect to the ionic strength of the test solution, amp/M/liter.

The ionic strength of the test solution can be determinated accurately for any given water by means of conductance measurements, as shown by the following equation

$$L = \theta + \lambda\ I - \delta I^2 + \Gamma\ I^3 \qquad (59)$$

where: L = Specific conductance, $\text{ohm}^{-1}\text{cm}^{-1}$

θ, λ, δ, and Γ = constants.

The nonlinear relationship given in equation (59) can be approximated, however, to:

$$\Delta L = K_i\ \Delta I \qquad (60)$$

where: K_i = Proportionality constant.

Equation (60) indicates the linear dependence of conductance on ionic strength, which holds in most surface waters, including estuarine water, but does not hold in certain industrial wastes of high salt content.

By combining equations (58) and (6), it follows that:

$$i = \phi_{o,s} \, e^{K'_s \, \Delta L} \, C \qquad (61)$$

where: $K_s = K_s K_i$

The exponential terms in equations (57) and (61) express the effect of the temperature and salt content. An electronic compensator[69] for changes in temperature and salinity, based on these equations, is shown in Figure 40.

Advantages - Membrane electrode systems offer the following advantages.

1. The activity (and concentration) of molecular oxygen can be determined by using membrane electrode systems. Activity determinations are highly significant whenever environmental measurements and biochemical systems are concerned.
2. Since the sensing element is separated from the test solution by a selective permeable membrane, it is possible to analyze for DO in solutions containing ionic and organic contaminants.
3. The membrane serves as a diffusion layer, the thickness of which is independent of the hydrodynamic properties of the test solution. Accordingly, membrane electrodes exhibit more stable performance characteristics in flowing solutions than conventional electrode systems.
4. Membrane electrode systems possess the unique ability of analysis for DO in aqueous, nonaqueous, and gaseous systems.
5. One of the main advantages of membrane electrode systems is their suitability for field use. Because of their ruggedness, portability, and ease of operation and maintenance, they are ideal instruments for the analysis of DO under adverse environmental conditions.
6. Being completely submersible, they can be used to analyze for DO in situ.
7. Like other electrochemical systems, membrane electrodes can be used for continuous monitoring.

Limitations - A DO determination by means of membrane electrodes is a relatively simple procedure; however, certain difficulties are sometimes encountered.

1. Most of the difficulties associated with these electrode systems are mainly caused by a lack of understanding of the procedure and principle of operation. In polarography the electrode surface is continuously renewed, but in membrane electrode systems this is not

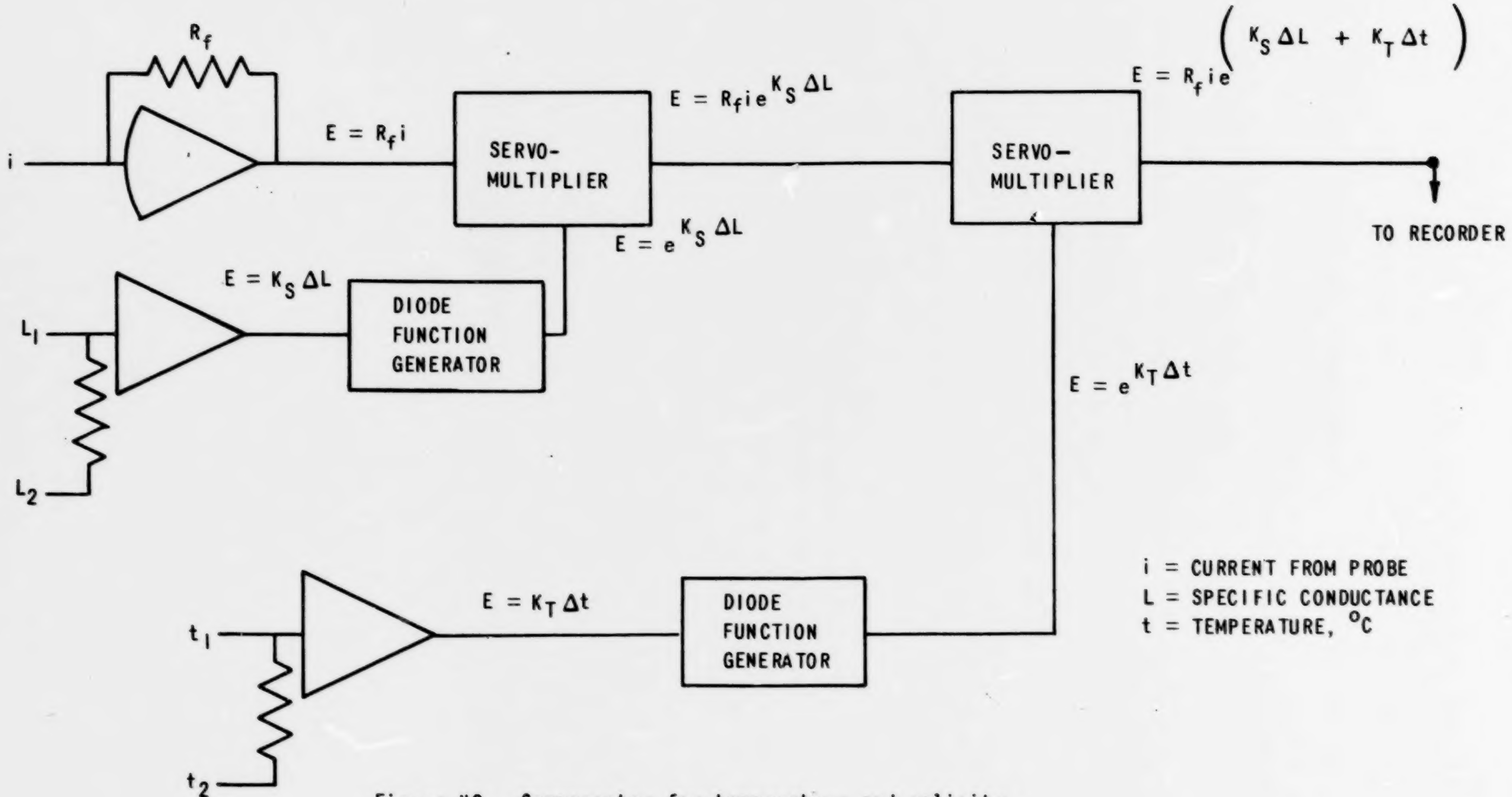

R_f
i
$E = R_f i$
SERVO-MULTIPLIER
$E = R_f i e^{K_S \Delta L}$
SERVO-MULTIPLIER
$E = R_f i e^{\left(K_S \Delta L + K_T \Delta t \right)}$
TO RECORDER
$E = e^{K_S \Delta L}$
L_1
L_2
$E = K_S \Delta L$
DIODE FUNCTION GENERATOR
$E = e^{K_T \Delta t}$
t_1
t_2
$E = K_T \Delta t$
DIODE FUNCTION GENERATOR
i = CURRENT FROM PROBE
L = SPECIFIC CONDUCTANCE
t = TEMPERATURE, °C

Figure 40. Compensator for temperature and salinity.

the case. Great care should be taken during the recharging of the electrode system to avoid contaminating the sensing element.

2. Improper mounting of the membrane might lead to trapping small air bubbles under the membrane. The bubbles cause a slower response to oxygen and a higher residual current.

3. Insufficient flow across the membrane surface results in erratic response; maintaining a threshold mixing value in the test solution is therefore essential.

4. Plastic membranes show selective permeability to various gases and vapors. Gases reduced at the potential of the sensing electrode, e.g., SO_2 and halogens, cause erroneous readings, but in aqueous systems these gases rarely exist in a free state. Other gases capable of permeating the plastic membrane may contaminate the sensing electrode or react with the supporting electrolyte, e.g., CO_2 and H_2S.

Applications - Oxygen membrane electrodes have been used in a variety of applications in the environmental sciences and engineering. Typical applications are oxygen monitoring in rivers and lakes, aeration studies, and oxygen utilization of domestic sewage.[70, 72, 121] Other applications include continuous DO recording in activated-sludge processes,[112] in respiration studies as a replacement of the Warburg Apparatus[65] in oceanographic studies,[24, 29] and in fermentation studies.[111]

IV. GENERAL DISCUSSION

CHOICE OF METHOD OF ANALYSIS

Chemical analysis for water quality characterization may be used to determine compliance with a given standard, to determine or forecast the effect of a water resource project, to evaluate treatment needs in view of the use of water, or to provide water quality control. These applications may require specific types of analysis with different levels of sophistication. It is unfortunate that the various methods of analysis for DO cannot be entered into a table that gives the suitable procedure to use for a given application. This is impossible primarily because in each application different problems will be encountered, both in terms of the characteristics of the water under test and in terms of the investigation requirements, e.g., analysis speed, frequency, and accuracy.

The analyst experienced in water quality characterization can often rely on his well-developed intuition to choose the appropriate method of analysis. In most cases, however, certain operating principles are needed for guidelines. Primarily the analyst should be familiar with the purpose of the analysis and the origin and properties of the water under investigation. Some of the questions for which answers are needed are:

1. What is the required degree of sensitivity and accuracy?
2. What should be the frequency and speed of analysis?
3. Will the analysis be performed in the laboratory or in the field?
4. What are the sampling procedures and sites?

Special attention should be given to in situ methods of analysis by which the species under test is determined in its own environment. In situ determination of DO is by far the most significant type of analysis for water quality characterization. Although electrochemical methods in general are capable of in situ analysis, the membrane electrode is the only system that is inherently suitable and that has been used successfully for in situ analysis in natural and waste waters.

CORRELATION OF DIFFERENT ANALYTICAL METHODS

Continuing technological advances in analytical chemistry and instrumentation during the past half-century have provided the chemist with a variety of methods of analysis for DO. Each of these methods utilizes a basic physical or chemical property of molecular oxygen. For example, in titrimetric and colorimetric methods the oxidizing property of oxygen is utilized; whereas, voltammetric methods depend on the rate of diffusion of molecular oxygen. Other methods of analysis are based on the determination of the thermal conductivity or the paramagnetic susceptibility of molecular oxygen.

Attempts to correlate results based on different methods of DO determination have caused a great amount of confusion, which has resulted in ambiguity as to how to define and express oxygen measurements. For example, medical scientists usually express oxygen measurements in blood or biological fluid as "oxygen tension"; oceanographers usually use the "partial pressure" scale; limnologists and aquatic biologists commonly use "percent saturation"; chemists use the molar concentration; and, last but not least, the sanitary engineer prefers "milligram per liter." In spite of the diversity of purposes behind the analysis and differences in the background of the analysts, the substance under test is the same, i.e., molecular oxygen.

To understand this point, one must realize that there is an inherent distinction: each method of analysis measures a different property of molecular oxygen. Since these properties are affected differently by changes in the physical and chemical properties of the environment, the results from various analytical methods may vary. For example, although changes in the temperature have slight or no effect on titrimetric methods of analysis, electrochemical methods such as conductometric, galvanic, voltammetric, and membrane electrodes are indeed temperature-sensitive. Recent investigations[69] have shown that changes in the ionic strength of the test solution do not interfere with titrimetric methods or with certain colorimetric methods, yet significantly influence electrochemical methods.

In the case of polarographic methods, changes in the ionic strength of the test solution influence the structure of the double layer at the dropping-mercury electrode and change the diffusivity coefficient of molecular oxygen.

INTENSIVE VERSUS EXTENSIVE MEASUREMENTS

By definition, extensive properties, such as volume and mass, are additive, in the sense that the value of the property for the whole of a system is the sum of the values for all of its constituent parts. On the other hand, intensive properties, such as pressure, density, and temperature, can be specified at each point in a system and may vary from point to point when there is an absence of equilibrium. In other words, intensive properties are not additive, and they do not give any specification of the quantity of the component they refer to in the system.

The chemical potential* is also an intensive property of the system. A difference of chemical potential may be regarded as the cause of a chemical reaction or of the tendency of a substance to diffuse from one phase to another. By definition, the chemical potential is related to the activity of a given component at constant temperature and pressure by the following equation:

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*See Appendix E for the definition and significance of the chemical potential.

$$\mu_i = \mu_i^o + RT \ln a \tag{63}$$

where μ is the chemical potential and a is the activity. The activity, sometimes referred to as the "effective concentration" or the "escaping tendency," is expressed as the product of the stoichiometric concentration, C, and the activity coefficient, γ, as shown by equation (7).

As far as chemical analysis is concerned, differentiation between intensive measurements is possible, depending upon whether the determined parameter is an intensive or an extensive property of the species under test. For example, titrimetric methods are essentially extensive measurements. They are based on the careful addition of a titrant to a reactive solution with the hope of locating the point of chemical equivalence upon which stoichiometric calculations may give the number of oxygen molecules per unit volume, i.e., the concentration.

On the other hand certain electrochemical methods are intensive measurements. This is best illustrated by the oxygen-sensitive membrane electrode where the current is solely dependent on the difference in the chemical potential across the membrane. In this case measurements are expressed in terms of the activity of molecular oxygen. Accordingly, if the membrane electrode is used to express the DO content of a certain water, this measurement does not have to be equal to results obtained by titration according to the Winkler Test. In the former case, the measured parameter is the activity of molecular oxygen; in the latter case, the number of oxygen molecules per unit volume (concentration) is determined. The same difference exists between pH measurements and acidity determinations. If a pH-type glass electrode is used to determine the hydrogen ion content of a water sample, the measurement may not be equal to results obtained by titration with a standard base. This example must be based on the assumption that the sample does not contain weak acids. The similarity between the pH-type glass electrode* and the oxygen membrane electrode is evident since in both systems the rate-determining step is the transport of the electroactive species through the membrane. This is solely dependent on the electrochemical potential difference, $\Delta\bar{\mu}$, across the membrane in the case of the glass electrode and the chemical potential difference, $\Delta\mu$, in oxygen membrane electrode systems. The electrochemical potential is by definition

$$\bar{\mu}_i = \mu_i + nFE \tag{64}$$

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*It is necessary to account for the liquid junction potential in order to determine the activity of hydrogen ions by a glass electrode.

Under ideal conditions, in the absence of interferences and extraneous environmental factors, the activity of molecular oxygen and its concentration are equal. In environmental systems it is unlikely for a variety of reasons that the two will exactly coincide, although in many applications the difference may be negligible for practical purposes.

SIGNIFICANCE OF ACTIVITY MEASUREMENTS

In spite of the fact that in most cases oxygen measurements are expressed in concentration units, the use of activity units is more significant, especially whenever environmental measurements are concerned. Perhaps the reader considers activity determination as too theoretical and of no value in practice. Let us consider, then, a typical case of natural or waste water in which molecular oxygen is involved in a variety of physical, chemical, and biological reactions. An example of the physical reactions is oxygen transfer across the air-water interface or within the bulk of the aqueous phase. Under constant temperature and hydrodynamic conditions, the driving force for diffusion of molecular oxygen is primarily proportional to the activity gradient and not necessarily to the concentration gradient. In other words, diffusion always occurs in the direction of falling activity and not always in the direction of diminishing concentrations. This analogy is similar to the flow of heat, which is always in the direction of falling temperature (temperature is an intensive parameter) and not in the direction of falling enthalpy per unit volume (an extensive parameter).

In view of the above discussion, under certain conditions, "uphill" diffusion may occur. Uphill diffusion, of course, means diffusion in the direction of increasing concentration, apparently counteracting an equalization of concentration. This is possible, if, for instance, a component not participating in diffusion causes a decrease in activity combined with an increase in concentration (salting-in effect) or an increase in activity combined with a decrease in concentration (salting-out effect).

On considering the chemical kinetics aspects of solution reactions, direct activity measurements are highly significant, especially in complicated systems such as natural or waste waters. Frequently, the chemist has had to calculate values of activity coefficients to account for environmental factors such as effect of pressure, ionic strength, solvent-solute or solute-solute interactions, and interferences caused by the actions of catalysts or inhibitors. From the point of view of chemical reactions, no generalization is possible, but it may be said for purposes of convenience that the overall rate can be either diffusion or activation-controlled. Most reactions in dilute aqueous systems fall in the former category, in which the rate-determining step involves a diffusion energy barrier. Under these conditions the reaction rate is directly dependent on the activity gradient of the reactive species. With regard to biological reactions, activity measurements offer distinct advantages over concentration determinations. In natural and waste waters, the availability of molecular oxygen to biological systems ordinarily depends primarily on the rate of oxygen transport. This can be either in the form of molecular diffusion in the aqueous phase or penetration through a biological membrane. In either case, the rate of oxygen uptake by the living organism can be correctly followed by the measurements of the activity rather than by the concentration of molecular oxygen.

How to measure the activity of molecular oxygen is also important to the reader. Oxygen-sensitive membrane electrode systems offer the most simple and direct method of activity determinations. Equation (55) shows that the steady-state current is directly proportional to the activity of DO. It is necessary here to adopt one convention that relates the activity to a certain concentration scale, as given by equation (7). The quantity of molecular oxygen in solution may be expressed as mole fraction, molality, or molarity. The mole-fraction scale has more theoretical significance than either molality or molarity. There are reasons for expecting that the partial pressure of a component is only proportional to its mole fraction, especially under simple molecular conditions, and this is one of the reasons that the mole fraction, rather than any alternative scale, is used for the definition of ideal solutions.

The molarity or volume concentration scale (e.g., mg/liter) is common and is easier to apply than the mole fraction. If V is the volume of the solution in liters, the molarity of the oxygen solution is defined by

$$C_i = n_i / V \tag{65}$$

where n_i = the number of gram-moles of the solute (oxygen). Let ρ be the density of the solution in g/ml; then the volume, V, is 1,000 ρV in grams and, therefore

$$1{,}000\ \rho V = M_o n_o + \Sigma M_i n_i \tag{66}$$

where: n_o = Number of gram-moles of the solvent (water)

M_o = Molecular weight of the solvent (water)

M_i = Molecular weight of the solute (oxygen).

Then,

$$V = \frac{M_o n_o + \Sigma M_i n_i}{1{,}000\ \rho} \tag{67}$$

and from (65), we obtain

$$C_1 = \frac{1{,}000\ \rho n_1}{M_o n_o + \Sigma M_i n} \tag{68}$$

Since by definition the mole fraction is equal to

$$X_i = \frac{n_i}{n_o + \Sigma n_i} \tag{69}$$

we obtain the following ratio:

$$\frac{C_i}{X_i} = \frac{1,000 \; \rho (n_o + \Sigma \; n_i)}{M_o n_o \; + \; \Sigma \; M_i n_i} \qquad (70)$$

As $\Sigma \; n_i \longrightarrow 0$, $C_i / X_i \longrightarrow 1,000 \; \rho / M_o$.

In very dilute solutions, therefore,

$$C_i = \frac{1,000 \; \rho \; X_i}{M_o} \qquad (71)$$

From equation (71), the molarity and mole fraction are directly proportional and can be used exchangeably in dilute solutions. As previously mentioned, since oxygen is a sparingly soluble gas, its solubility in water follows closely Henry's Law. Under 1 atmosphere of air, therefore, the molar concentration can be used instead of the mole fraction to give the following form of Henry's Law.

$$P' = H' \gamma \; C = H \; a \qquad (72)$$

where: H = Henry's Law constant on a molarity scale

γ = Activity coefficient

C = Molar concentration of DO

a = Activity of DO, molar scale

P_i = Oxygen partial pressure in 1 atmosphere of air, atm.

Under these conditions the activity coefficient, γ, is equal to unity, and the activity and concentration of molecular oxygen are equal. Accordingly, the membrane electrode can be easily calibrated for activity determinations by using a sample of distilled water in equilibrium with air and measuring the diffusion current and the concentration of DO by the Winkler Test. The concentration value is taken in this case to be numerically equal to the oxygen activity in the water sample and to be directly proportional to the diffusion current as shown by equation (55).

In view of the foregoing discussion, direct measurement of the activity of molecular oxygen in natural and waste waters is highly significant and is relatively simple. In situ measurement of DO activity by membrane electrodes is not widely practiced. Once it is adopted, a rich harvest of answers to some of the complicated biochemical aspects of the aquatic environment should be forthcoming.

V. NOTATIONS

A = Thickness of the electrolyte solution film, cm

a = Activity, molar scale

$\bar{a}$ = Surface area of indicator electrode, cm^2

B = Thickness of the membrane layer, cm

C = Concentration of dissolved oxygen, M/liter

C_s = Salt concentration, mg/liter

D = Diffusivity coefficient, cm^2/sec

E_m = Measured electrode potential, v

E_o = Standard electrode potential, v

E_p = Activation energy of permeation of membrane, cal/M

F = The Faraday

G = Free energy

g = Acceleration, cm/sec^2

g = Weight of substance electrolyzed

H = Henry's Law constant, 5.89×10^5 mm Hg/M/liter

h = Depth, cm

I = Ionic strength

i = Current, amp

i_d = Diffusion current, amp

J = Flux of molecular oxygen, $moles/cm^2$-sec

K_i = Conductance proportionality constant

K_s = Modified salting-out coefficient

K_s = Salting-out coefficient

L = Specific conductance, ohm^{-1} cm^{-1}

M = Molecular weight, g/mole

m = Rate of flow of mercury, mg/sec

m^o = Temperature coefficient, ^oK

n = Number of electrons transferred in oxidation-reduction process per mole

N_i = Number of gram moles of solute

P = Partial pressure of oxygen, 154 mm Hg at sea level

P_m = Permeability coefficient, cm^2/sec

q = Number of coulombs passed in electrolysis

R = Gas constant = 0.0821 L -atm/M/^oK

S = Oxygen solubility, mg/liter

S_o = Oxygen solubility in distilled water, mg/liter

T = Absolute temperature, ^oK

T_c = Temperature, ^oC

t = Time

V = Volume

v = Velocity of molecular oxygen, cm/sec

$\bar{v}$ = Partial molal volume of solute, liter/atm

X = Mole fraction

x = Distance, cm

Z = Ionic charge

α = Constant

β = Constant

Γ = Constant

λ = Constant

δ = Constant

γ = Activity coefficient

ρ = Density of solution, g/cm^3

ΔH = Heat of solution, cal/M

ψ = Electrostatic charge

μ = Chemical potential, cal/M

$\bar{\mu}$ = Electrochemical potential

θ = Constant

ϕ = Sensitivity coefficient, μa per unit concentration

VI. REFERENCES

1. Alcock, G. P., and K. B. Coates. Indigo-carmine for the colorimetric determination of low concentrations of dissolved oxygen in water. Chemistry and Industry. No. 19, 554. 1958.

2. Aleshovshii, V. B., and V. A. Kovalstov. Quantitative determination of oxygen dissolved water. USSR Pat. 144, 046. 1962.

3. Alsterberg, G. Methods for the determination of elementary oxygen dissolved in water in the presence of nitrites. Biochem. Z. 159:36. 1925.

4. Ambuhl, H. Electrochemical method of determination of oxygen in water. I. Principles; long-term measurements. Schweiz. Z. Hydrol. 17:123. 1955.

5. Ambuhl, H. Practical application of the electrochemical determination of oxygen in water. Schweiz. Z. Hydrol. 20:341. 1958.

6. Ambuhl, H. Practical application of the electrochemical determination of oxygen in water. Schweiz. Z. Hydrol. 22:23. 1960.

7. Arnott, J., and J. McPheat. Estimation of dissolved oxygen in deaerated water. J. Engineering. 176:103. 1953.

8. ASTM, Manual on industrial water and industrial waste water. 2nd ed. 1962.

9. ASTM Standards, Part 7, 1155-62. 1952.

10. Bairstow, S., J. Francis, and G. H. Wyatt. Photometric determination of dissolved oxygen in condensates and feed waters by means of starch-iodide complex. Analyst. 72:340. 1947.

11. Ballinger, D. Polarographic methods for water and wastes. JWPCF. 35:116. 1963.

12. Banks, J. Absorptiometric determination of dissolved oxygen. Analyst. 79:170. 1954.

13. Barbi, G., and S. Sandroni. Internal coulometric methods for the determination of oxygen dissolved in water. Comit. Nazl. Energia Nucleare. CNI-55, 3. 1960.

14. Barhin, M. B. Use of solid rotating platinum disk microelectrode in

polarographic analysis. Uchenye Zapishi. Kishinev Gosudarst Univ., 56:99. 1960.

15. Beckman Instrument, Inc. Continuous oxygen analyzers. Bulletin 0-4016, Beckman Instrument, Inc. Fullerton, Calif.

16. The Beckman Laboratory Oxygen Sensor. Bulletin 7013. Beckman Instruments, Inc. Fullerton, Calif.

17. Briggs, R., F.S. Daview, G.V. Dyke, and G. Knowles. Use of wide-borne dropping mercury electrode and zinc reference electrode for continuous polarography. Chemistry and Industry, No. 8, 223. 1957.

18. Briggs, R., G.V. Dyke, and G. Knowles. Use of the wide-borne dropping mercury electrode for long-period recording of concentrations of dissolved oxygen. Analyst. 83:304. 1958.

19. Briggs, R., and N.H. Mason. A portable dissolved oxygen indicator. Lab Pract. 11:36. 1962.

20. British Standards Institution. 1427, 1949. 2690, 1956.

21. Buchoff, L.S., N.M. Ingber, and J.H. Brady. Colorimetric determination of low concentrations of dissolved oxygen in water. Anal. Chem. 27:1401, 1955.

22. Butler, R.A., J.F. Nuna, and S. Ashill. Coiled cathode oxygen polarograph. Nature. 196:97, 1962.

23. Carritt, D.E., and J.W. Kanwisher. An electrode system for measuring dissolved oxygen. Anal. Chem. 31:5, 1959.

24. Cassie, R.M. Calibration and operation of dissolved oxygen electrode. New Zealand Oceanographic Institute. 1963.

25. Cleary, E.J. An electronic monitor system for river quality surveillance and research. Proc. 1st Intern. Conf. on Water Poll. Res., Pergamon Press. 1962.

26. Cleary, E.J. Introducing the ORSANCO robot monitor. Water Quality Measurement and Instrumentation. Proc. USPHS Symposium, p. 108. 1962.

27. Daynes, H.A. Gas analysis by measurement of thermal conductance. Cambridge University Press. London. 1933.

28. Dixon, W.S. Pollution control by continuous dissolved oxygen analysis and associated instruments. Proc. Water Quantity Measurement and Instrumentation. Technical Report, Robert A. Taft Sanitary Engineering Center, Cincinnati, Ohio. 1960.

29. Duxbury, A.C. Calibration and use of a galvanic type oxygen electrode in field work. Limnol. and Oceanography. 8:483. 1963.

30. Dykes, D.F., and J.C. Baumgartner. Use of pure oxygen for aerating craft mill wastes. Proc. 6th Southern Munic. and Ind. Wastes Conf., N. C. State Coll. p. 200. 1957.

31. Eckenfelder, J. W.W., and C.T. Burris. Polarographic measurement of dissolved oxygen. ASTM Special Technical Publication. 219:18. 1957.

32. Eckfeldt, E.L., and E.W. Shaffer. Dissolved oxygen measurement by constant-potential derivative coulometry. Anal. Chem. 36:2008. 1964.

33. Erdey, L., and F. Szabadvary. Ascorbometric determination of oxygen dissolved in water. Acta. Chem. Acad. Sci., Hungary, 4·325. 1954.

34. Finnegan, T., and R. Tucker. The beckman dissolved oxygen analyzer. ASTM Special Technical Publication No. 219. 1959.

35. Fitts, D.D. Nonequilibrium thermodynamics. McGraw-Hill. New York. 1962. p. 83.

36. Freier, R., and G.Z. Resch. Determination of the dissolved oxygen in water. Anal. Chem. 148:427. 1956.

37. Galvanic cell oxygen analyzer. Precision Scientific Co., 3737 W. Cortland St. Chicago 27, Ill.

38. G.M.E. Oxygraph. Bulletin, Gilson Medical Electronics, Middleton, Wis.

39. Garst, A.W., and T.W. McSpadden. Measurements of dissolved oxygen in oil field waters. Pan American Petroleum Corporation, Tulsa, Okla. 1963.

40. Grasshoff, K. Determination of oxygen in sea water. Kiel Meeresforsch. 18:42. 1962.

41. Hays thermal conductivity analyzer. The Hays Corporation. Michigan City, Ind. 1960.

42. Hays Magno-Therm oxygen recorder. The Hays Corporation. Michigan City, Ind. 1960.

43. Hersh, P.A. Galvanic Analysis. In: Advances in analytical chemistry and instrumentation, Vol. III. C.N. Reilly, ed. Academic Press. 1964. p. 210.

44. Hissel, J., and J. Prie. Microdetermination of dissolved oxygen in boiler feed water by coulometry at constant current. Bull. Centre Belge Etude et Document Eaux. 44:76. 1959.

45. Inczedy, J. Redox reaction on ion exchange columns. II. Reduction and oxidation of some inorganic and organic substances for analytical purposes. Acta. Chim. Acad. Sci. Hungary. 27:185. 1961.

46. Ingols, R. S. Determination of dissolved oxygen by the dropping mercury electrode. Sew. Works J. 13:1097. 1951.

47. Interstate Commission on Potomac River Basin "News Letter". 18: 7. 1962.

48. James, G. S., and M. H. Stephan. The electrometric determination of dissolved oxygen in aqueous solution. Analyst. 85:35. 1960.

49. Jarrel Ash Company. Dissolved oxygen analyzer. Special bulletin. Boston, Mass. 1962.

50. Johnson, F. D. Development of field test for free chlorine. Department of Environmental Health, University of North Carolina. Chapel Hill, N. C. 1964.

51. Johnson, M. J., J. Borkowski, and C. Engblom. Stream sterilizable probes for dissolved oxygen measurements. Biotechnology and Bioengineering. VI. 457. 1964.

52. Karchmer, J. H. Polarographic determination of dissolved oxygen-study of drop time with rapid dropping electrode. Anal. Chem. 31: 502. 1959.

53. Keidel, F. H. Coulometric analyzer for trace quantities of oxygen. Ind. Eng. Chem. 52:490. 1960.

54. Klima, D. Simple arrangement for the polarographic determination of dissolved oxygen in industrial wastes. Voda. 36:148. 1957.

55. Klotz, I. M. Variation of solubility with depth. Limnol. Oceanog. 8:149. 1963.

56. Knowles, G., and G. F. Lowden. Methods for detecting the endpoint in titration of iodine with thiosulphate. Analyst. 78:159. 1953.

57. La Mout, B. D., and N. E. Gordon, Jr. Spectrophotometric determination of dissolved oxygen in water. USAEC. WCAP-917. 1953. 13 pp.

58. Lederer, V. V. Cerimetric determination of oxygen dissolved in water. Chem. Listy. 41:230. 1947.

59. Levin, H.S., and R.S. Kleinschmidt. Principle and problems in development of a dissolved oxygen analyzer. Sewage and Ind. Wastes. 29:856. 1957.

60. Levine, H.S., et al. Continuous measurement of dissolved oxygen in water. Anal. Chem. 28:343. 1956.

61. Lewin, V.H., and G.J. Phillips. Potentialities of polarographic techniques for sewage works analysis. Inst. Sewage Purif. J. Proc. 473. 1960.

62. Lingane, J.J. Chronopotentiometric study of oxygen reduction at a platinum wire cathode. J. Electroanal. Chem. 2:296. 1961.

63. Lipner, H.J., L.R. Witherspoon, and W.C. Champeaux. Adaptation of a galvanic cell for microanalysis of oxygen. Anal. Chem. 36:204. 1964.

64. Loomis, W.F. Improved rapid colorimetric microdetermination of dissolved oxygen. Anal. Chem. 28:1347. 1956.

65. Loomis, W.F. Rapid microcolorimetric determination of dissolved oxygen. Anal. Chem. 26:402. 1954.

66. Lynn, W.R., and D.A. Okun. Experience with solid platinum electrodes in the determination of dissolved oxygen. Sew. and Ind. Wastes. 27:4. 1955.

67. Macklin, M.O., D.J. Baumgartner, and M.B. Ettinger. Performance test of continuous recording dissolved oxygen analyzer. Sew. and Ind. Waste. 31:804. 1959.

68. Mancy, K.H., and D.A. Okun. Automatic recording of dissolved oxygen in aqueous systems containing surface active agents. Anal. Chem. 32:108. 1960.

69. Mancy, K.H., and J.E. Haskins. The voltammetric analysis of dissolved oxygen. I. the salt effect. 148th ACS Meeting, Chicago. Sept. 1964.

70. Mancy, K.H., and D.A. Okun. Oxygen in waste treatment - the galvanic cell oxygen analyzer. special report to NIH, Dept. of Environmental Sciences and Engineering, School of Public Health, University of North Carolina, Chapel Hill, N.C. 1961.

71. Mancy, K.H., D.A. Okun, and C.N. Reilly. The galvanic cell oxygen analyzer. J. Electroanal. Chem. 4:65. 1962.

72. Mancy, K.H., and W.C. Westgarth. A galvanic cell oxygen analyzer. JWPCF. 34:1037. 1962.

73. Manning, M. A method of obtaining continuous recording of dissolved oxygen in lake waters. Ecology. 21:509. 1940.

74. McKeown, J.J., G.W. Gore, and L.C. Brown. Comparative studies on the analysis of dissolved oxygen. National Council for Stream Improvement, Inc. Tufts Univ. Medford, Mass. 1964.

75. McCrumb, F.R., and W.R. Kenny. New method for determining small amounts of dissolved oxygen. JAWWA. 21:400. 1929.

76. Meyer, H.J., and C. Brack. Simplified determination of dissolved oxygen in water by means of o-tolidine. Chem. Eng. Tech. 22:545. 1950.

77. Meyling, A.H., and G.H. Frank. A simple method for determining dissolved oxygen with indigo carmine. Analyst. 87:60. 1962.

78. Moore, E.W., J.C. Morris, and D.A. Okun. The polarographic determination of dissolved oxygen in water and sewage. Sew. Works J. 20:1041. 1948.

79. Morgan, J.J., and W. Stummn. The role of multivalent metal oxides in limnological transformations, as exemplified by iron and manganese. Preprint, Second International Conference on Water Pollution Research. 1964.

80. Morris, J.C. Final report, Contract SAph 69705. Department of Health, Education, and Welfare with Harvard University. May 1, 1958 to January 31, 1959.

81. Nakatomi, K. Colorimetric method for determining dissolved oxygen in sea water. Koggo Kazaku Zasshi. 62:503. 1959.

82. Ohle, W. Die chemische und die elektrochemische bestimmung des molekular gelosten sauerstoffs der binnengewasser. Int. Ver. f. Theoror, u. Angew. Limnologie. Mitteilungen. 3:44. 1953.

83. Okun, D.A., J.C. Lamb, and C.C. Wells. A waste control program for a river with highly variable flow. Proc. 17th Purdue Ind. Waste Conf. 1962.

84. Okura, T., K. Goto, and K. Furukawa. Determination of dissolved oxygen in polluted waters. Mem. Fac. Engr. Hokkaido University. 11:11. 1960.

85. Parker, B.W. Automatic system for monitoring water quality. J. San. Engr. Div. Proc. ASCE, SA 4, 25. 1960.

86. Paris, J.P., J.D. Gorsuch, and D.M. Hercules. Titration of oxygen and antioxidants using 2, 4, 6-tri-tetra-butylphenoxy free radicals. Anal. Chem. 36:1332. 1964.

87. Pieters, H.A.J., and W.J. Hanssen. Spectrophotometric determination of traces of oxygen in water. Anal. Chim. Acta. 2:712. 1948.

88. Pomeroy, R., and H.D. Kirschman. Determinations of dissolved oxygen-proposed modification of the Winkler Test. Anal. Chem. 17:715. 1945.

89. Potter, E.C., and G.E. Everitt. Advances in dissolved oxygen microanalysis. J. Appl. Chem. 9:642. 1959.

90. Potter, E.C. Microdetermination of dissolved oxygen in water. I. Nature of the problem. J. Applied Chem. 7:285. 1957.

91. Potter, E.C., and T.F. White. J. Applied Chem. London. 7:309. 1957.

92. Pytokowicz, R.M. Comment on the variation of solubility with depth. Limnol. Oceanog. 8:486. 1963.

93. Rand, C.M., and H. Heukelekian. Determination of dissolved oxygen in industrial wastes by the Winkler and polarographic methods. Sew. Ind. Wastes. 23:1141. 1951.

94. Richter, H.G., and A.S. Gillespie, Jr. Thallium-204 radiometric determination of dissolved oxygen in water. Anal. Chem. 34:1116. 1962.

95. Rideal, S., and G.G. Stewart. The determination of dissolved oxygen in water in presence of nitrites and organic matter. Analyst. 26:141. 1901.

96. Ruchhoft, C.C., and W.A. Moore. The determination of the biochemical oxygen demand and dissolved oxygen of river mud suspensions. Ind. Eng. Chem. 12:711. 1940.

97. Ruchhoft, C.C., and O.R. Placak. Determination of dissolved oxygen in activated-sludge sewage mixtures. Sew. Works J. 14: 638. 1942.

98. Sawyer, D.T., R.S. George, and R.C. Rhodes. Polarography of gases - quantitative studies of oxygen and sulfur dioxide. Anal. Chem. 31:2. 1959.

99. Sawyer, D.T., and L.V. Interrante. Electrochemistry of dissolved gases. II. Reduction of oxygen at platinum, palladium, nickel, and other metal electrodes. J. Electroanal. Chem. 2:310. 1960.

100. Scholander, D.L., et al. Micro-gasometric determination of dissolved oxygen and nitrogen. Biol. Bull., Woods Hole, 109:328. 1955.

101. Seaman, W., and W. Allen. Polarographic determination of dissolved oxygen in water containing industrial effluents. Sew. Ind. Wastes. 23:1141. 1951.

102. Solubility of atmospheric oxygen. Twenty-ninth progress report of the Committee on Sanitary Engineering Research of the Sanitary Engineering Division of ASCE, Proc. ASCE, 86:41. 1960.

103. Splittgerber, A., et al. Determination of dissolved oxygen in water by means of o-tolidine. Chem. Eng. Tech. 22:542. 1950.

104. Spoor, W.A. Use of the dropping mercury electrode for the continuous measurement of dissolved oxygen in flowing water. Science. 108:421. 1948.

105. Standard methods for the examination of water and wastewater. APHA. AWWA. WPCF. 10th ed. 1955.

106. Standard methods for the examination of water and wastewater. APHA. AWWA. WPCF. 11th ed. 1960. p. 392.

107. Stone, H.W., and R.L. Eichelberger. Titration of dissolved oxygen with acid-chromous reagent. Anal. Chem. 23:868. 1951.

108. Stones, W.F. The use of hydrazine as an oxygen scavenger in H.P. boilers. Chem. and Ind. 6:120. 1957.

109. Stracke, Gerhard. Continuous measurement of dissolved oxygen. Vom. Wasser. 25:181. 1958.

110. Strefelda, F. Polarography in flowing electrolytes. Collection Czechoslov. Chem. Communs. 25:862. 1960.

111. Strohm, J.D., and R.F. Dale. Dissolved oxygen measurement in yeast propagation. Ind. Eng. Chem. 53:760. 1961.

112. Sulzer, F., and W.M. Westgarth. Continuous DO recording in activated sludge. Water and Sewage Works. 8:266. Oct. 1962.

113. Thayer, L.C., and D.A. Robinson An instrument for the continuous determination of dissolved oxygen based on paramagnetism. First International Congress of Instrumental Society of America. Philadelphia. V. 9, P. 2, paper 54-22-4, 1954.

114. Theriault, E.G., and P.D. McNamee. Dissolved oxygen determination in the presence of organic matter, hypochlorites and sulfite wastes. Ind. Eng. Chem. 4:59. 1932.

115. Todt, F. Application of the electrochemical determination of oxygen. Angewandte Chemie. 9:266. 1955.

116. Todt, F. Electrochemische Sauerstoffmessungen. W. de Gruyter and Co. Berlin, 1958. Dechema Monographien. 35:132. 1959.

117. Todt, F., and R. Freier. Electrochemical determination of oxygen in water. Ger. Pat. 1,012,764. 1957.

118. Toller, W. Methods of determining oxygen dissolved in boiler feed water. Mitt Ver Grosskesselbes. 16:416. 1951.

119. Tyler, C.P. Dissolved oxygen determination. U.S. Pat. 2,993,846. July 25, 1961.

120. Tyler, C.P., and J.H. Karchmer. Portable analyzer for determination of dissolved oxygen in water - application of rapid-dropping mercury electrode. Anal. Chem. 31:499. 1959.

121. Weiss, C.M., and R.T. Oglesby. Instrumentation for monitoring water quality in reservoirs. Presented at 83rd Annual Conference of AWWA, May 23, 1963.

122. Whitney, R.J. A syringe pipette method for the determination of oxygen in the field. J. Exp. Biol. 15:564. 1938.

123. Williams, D.D. and R.R. Miler. An instrument for on-stream stripping and gas chromatographic determination of dissolved gases in liquids. Anal. Chem. 34:657. 1962.

124. Wilson, Ray I., and Jan N. Haagen-Smit. Measurement of dissolved oxygen. U.S. Pat. 3,042,495. July 3, 1962.

125. Winkler, L.W. The determination of dissolved oxygen in water. Ber. Deut. Chem. Ges. 21:2843. 1888.

126. Wright, J.M., and W.T. Lindsay, Jr. New method for the continuous analysis of dissolved oxygen in water. Proceedings of Am. Power Conf. 21:706. 1959.

127. Yakimets, E.M., and N.U. Shabanova. Determination of oxygen dissolved in water in presence of nitrites. Truday Ural Politekh Inst. Sbornik. 57:79. 1956.

128. Zimmerman, M. Mitt. Ver. Groesskesselbesitz. 6:35. 1949.

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VIII. APPENDICES

APPENDIX A: WINKLER REACTIONS AND IODINE-THIOSULFATE TITRATION

The principal reactions involved in the Winkler Test are:

1. The quantitative oxidation of alkaline manganous hydroxide by the oxygen contained in the water sample.

$$Mn^{2+} + 2OH^- \longrightarrow Mn(OH)_2 \text{ (White precipitate)} \qquad (A\text{-}1)$$

$$Mn(OH)_2 + 1/2O_2 \longrightarrow MnO_2 \cdot H_2O \text{ (Brown precipitate)*} \qquad (A\text{-}2)$$

2. Oxidation of iodide ions by the manganic salt after acidification, with the release of free iodine.

$$MnO_2 \cdot H_2O + 2I^- + 4H^+ \longrightarrow Mn^{2+} + I_2 + 3H_2O \qquad (A\text{-}3)$$

The manganese ions function as an oxygen carrier or temporary fixative, and the amount of iodine liberated is equivalent to the oxygen originally present in the test solution.

3. The quantitative determination of the released iodine by one of several methods of iodometric analysis. In iodometric analysis, accuracy depends mainly on the technique used.

In the standard Winkler Test,[106] the liberated iodine is titrated directly with a standard thiosulfate solution, as follows:

$$I_2 + 2S_2O_3^{2-} \longrightarrow S_4O_6^{2-} + 2I^- \qquad (A\text{-}4)$$

The end point is usually detected visually by means of starch as an indicator. The complex compound formed by the interaction of iodine on starch with its characteristic blue color, is capable of indicating the presence of molecular iodine as low as 10^{-5} M.

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*Recent investigation on the chemistry of manganese revealed that the reaction product in equation (A-2) is not manganese oxide but rather a mixed oxide.[79]

Since iodine reacts with excess iodide ions to form I_3^-, the actual reaction that occurs when an acidic solution of iodine in potassium iodide is titrated with thiosulfate is as follows:

$$I_3^- + 2S_2O_3^{2-} \longrightarrow 3I^- + S_4O_6^{2-} \qquad \text{(A-5)}$$

An intermediate ion, $S_2O_3I^-$, is formed, however, by the reversible reaction

$$S_2O_3^{2-} + I_3^- \rightleftharpoons S_2O_3I^- + 2I^- \qquad \text{(A-6)}$$

This intermediate may react with either I_3^- or iodide

$$2S_2O_3I^- + I^- \longrightarrow S_4O_6^{2-} + I_3^- \qquad \text{(A-7)}$$

$$S_2O_3I^- + S_2O_3^{2-} \longrightarrow S_4O_6^{2-} + I^- \qquad \text{(A-8)}$$

Equation (A-7) accounts for the reappearance of iodine near the end point in the titration of very dilute iodine solutions. The square of the concentration of the intermediate ion, $S_2O_3I^{2-}$, determines the rate of the overall reaction. The stoichiometry is no longer exact at low concentrations of iodide (below 0.003 M) because of some sulfate formation from the

$$S_2O_3I^- + 3I_3^- + 5H_2O \longrightarrow 2SO_4^{2-} + 10H^+ + 10I^- \qquad \text{(A-9)}$$

Since the rate of this latter reaction involves the first power of the $S_2O_3I^-$ concentration, it becomes more important toward the end of the titration as the $S_2O_3I^-$ concentration becomes smaller.

In alkaline solution ($pH > 8$) iodine reacts with hydroxyl ions; therefore, to insure proper stoichiometry, a pH of less than 5 is desirable. In an extremely acid solution, however, decomposition of thiosulfate may result. Acidification should therefore be kept to a minimum.

APPENDIX B - MICRO-WINKLER METHOD

In the micro method a syringe is used as the reaction vessel. A 1- to 2-ml water sample and the Winkler reagents are taken into the syringe, allowed to react to liberate iodine, and then expelled for micro-titration. For calibration of the syringe, the total volume (barrel plus dead space in the nozzle of the syringe) should be accurately determined. When the test is performed, the dead space of the syringe is first filled with manganous chloride solution. This is followed by drawing in the water sample to mix the sample with the manganous chloride. Alkaline iodide solution is then drawn into the syringe, which is shaken until the precipitate is evenly distributed. The syringe is then laid on its side for 3 minutes, in the dark, to allow completion of the reaction with oxygen. This is followed by drawing in phosphoric acid and shaking by inversion until all the precipitate has dissolved. The iodine thus liberated and the washings are then ejected into the titration vessel. A micro-burette is used to titrate the iodine with a 0.005N standard thiosulfate solution.

APPENDIX C - PROCEDURE FOR GASOMETRIC METHOD

The gasometric apparatus as reported by McKeown et al.[74] is illustrated in Figure C-1. It consists mainly of a mercury leveling bottle, 500-ml evacuation vessel, 15-ml reaction chamber, and two-way stopcocks.

A. Introductory Notes

1. Prior to a test, the stopcocks and ground-glass ball joints must be carefully lubricated.

2. Approximately 28 lb of Hg is introduced into the leveling bottle. One in. of distilled water is placed on top of the mercury, and 3 to 4 ml of water is also added to the evacuation vessel.

3. In all manipulations, it is advisable to check the upward rush of mercury by pinching the rubber tubing.

4. The stopcocks must be open to allow air to escape when introducing Hg; otherwise a closed system is created, and the reaction chamber may be ruptured.

5. All tubing should be securely wired to the bottles.

B. Test for Leakage

1. With the sampling port open to the atmosphere, the vacuum chamber is filled with Hg by raising the leveling bottle. Allow the sampling port to fill with water.

2. When the sampling port is filled with water, the lower stopcock is turned and the Hg is allowed to fill the reaction chamber with the upper stopcock open to the atmosphere (position shown in Figure C-1).

3. The Hg is allowed to rise in the reaction chamber until all of the water is in the stopcock and the Hg level just reaches the stopcock.

   Close the upper stopcock. Mark the position of the leveling bottle.

4. Slowly lower the leveling bottle to about 3 ft below the vacuum chamber. A torricellean vacuum is now created. The Hg should not go below the top of the outlet in the vacuum chamber.

5. Allow the Hg to rise again in the vacuum and reaction chambers by raising the leveling bottle to the position recorded

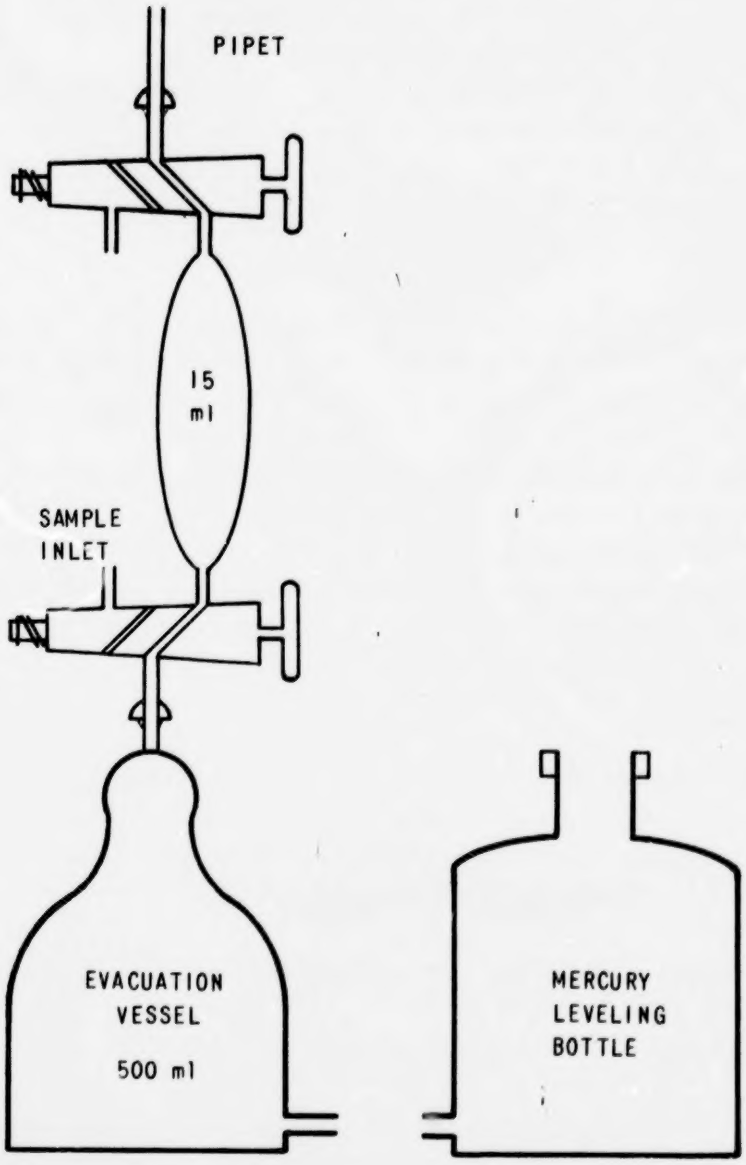

Figure C-1. Gasometric apparatus.

in step B-3 above. The Hg should again fill the chambers to the bottom of the upper stopcock in the absence of leakage. If air bubbles form in the water covering the Hg, exhaust the air and repeat the operation until the Hg returns to its original level.

6. Leakage may be localized by separate tests of the vacuum chamber and the reaction chamber.

C. Testing a Sample

1. With the apparatus in the position of step B-4 so that a vacuum exists in the vacuum chamber, a 75- to 100-ml sample is introduced through the sampling port by suitable manipulation of the stopcock. The sample should be flowing through the siphon (the siphon should be purged), and the sampling port should be filled when the sample is in-

troduced to prevent entrainment of air.

2. The sample is stirred in the vacuum chamber for 10 minutes to allow extraction of all gases. Stirring is accomplished by means of a magnetic mixer and a small stirring bar, which should be placed in the vacuum chamber prior to filling with Hg.

3. After the extraction has been completed, the gases are transferred to the reaction chamber by opening the stopcock to the reaction chamber and raising the leveling bottle.

4. When the sample reaches the top of the lower stopcock, the stopcock is turned so that the sample is allowed to run out of the sampling port into a graduated cylinder for determination of volume. Suitable allowances after calibration may be made for sample remaining in the lower stopcock.

D. Gas Analysis

1. The standard Winkler chemicals (Modification A) are used for the oxygen analysis. The chemicals are introduced with the small pipet provided. Before each chemical is introduced, one pipet full should be run to waste.

2. Two ml of $MnSO_4$ are introduced to the reaction chamber.

3. Rinse and add 2 ml of standard KI solution.

4. The vacuum remaining in the reaction chamber is broken by admitting 4 to 5 ml of distilled water of known DO content from the calibrated pipet.

5. The reaction chamber is removed from the apparatus and placed in a mechanical shaker (Eberbach 6460 Kahn Shaker 280 rpm) for 20 minutes, at which time the absorption of oxygen should be completed.

6. The reaction chamber is put back on the gasometric apparatus, and the floc is dissolved by introducing 2 ml of concentrated sulfuric acid by the alternative application of suction and pressure.

   CAUTION: Use a clip to hold the pipet in place, and wear safety glasses.

7. The iodine solution with rinsings and distilled water are transferred to a titration vessel to a volume of 200 ml.

8. The solution is then titrated with 0.025N sodium thiosulfate as in the standard method.

E. Calculations - Example

Assume a 100-ml sample:

A = 0.25N thiosulfate required for titration = 1.75 ml

B = Total oxygen = 0.2A = 0.350 mg

C = Correction for distilled water added to break vacuum assuming 4 ml used with DO = 8.0 mg/liter

$$\frac{4 \times 8.0}{1,000} = 0.032$$

D = Oxygen extracted from sample = B - C = 0.318 mg

E = Oxygen content = $\frac{1,000 \text{ ml}}{\text{vol. sample}} \times$ D = 3.18 mg/liter.

APPENDIX D - MICRO-GASOMETRIC METHOD

In the micro-gasometric method a 1-ml water sample is drawn into a special syringe pipet (Figure 18) containing sodium bicarbonate solution in the dead space in the nozzle of the syringe. Following this, citric acid is drawn in and the pipet is closed so that the carbon dioxide generated by the action of the acid on the bicarbonate will remain as a bubble in the solution. This relatively large volume of carbon dioxide will strip out all the dissolved gases originally present in the sample when the pipet is shaken. After ejecting the liquids from the syringe and yet retaining all the gases, a small volume of concentrated potassium hydroxide is drawn into the syringe. This absorbs the carbon dioxide and leaves a bubble of oxygen and nitrogen only. The bubble is then transferred to the bubble analyzer (Figure 18), which has a calibrated capillary tube for measuring the volume of gas. After the total oxygen and nitrogen volume is measured, a strong pyrogallol solution is introduced into the analyzer. This absorbs the oxygen from the bubble and leaves the nitrogen. The new volume, therefore, is that due to nitrogen, and the difference between the volumes is due to the oxygen in the original sample.

APPENDIX E - CHEMICAL POTENTIAL

The word "potential" is commonly used in mechanical or electrical systems. A potential has associated with it a capacity quantity, and the work done on a system is given by the product of the capacity factor times the change in potential. Thus for gravitational potential, $\bar{g}h$, the mass, m, is the capacity factor, and the work is $m\bar{g}(h_2 - h_1)$ where h is the elevation. For electrostatic systems the charge, φ, is the capacity factor, and the electrostatic potential is designated E. The work required to move a charge, φ, from a location of potential E_1 to another potential, E_2, is $\varphi(E_2 - E_1)$.

With chemical or biochemical systems, potential and capacity quantities can also be defined. If we choose the mole as the capacity factor and deal with systems at constant temperature and pressure, then the "free energy per mole" or the "molar free energy" is clearly the "chemical potential." The chemical potential, μ, is defined for molecular oxygen by the following equation:

$$\mu_{O_2} = \left(\frac{\partial G}{\partial n_{O_2}}\right)_{P,T,n_j} \qquad \text{(E-1)}$$

where: G = Free energy

n_{O_2} = Number of moles of molecular oxygen

n_j = Number of moles of all other components in the system

P = Pressure

T = Absolute temperature, ^{o}K

The activity of molecular oxygen, a_{O_2}, is related to the chemical potential by the following equation:

$$\mu_{O_2} = \mu^{o}_{O_2} + RT \ln a_{O_2} \qquad \text{(E-2)}$$

where: $\mu^{o}_{O_2}$ = A constant, which depends upon an arbitrarily chosen standard state of the component O_2 and upon the temperature and pressure

R = Gas constant.

In any system at equilibrium the chemical potential, μ_{O_2}, of component O_2 is constant throughout the system. If μ for any component varies from one region of a system to another, then the component will flow from the region of high μ toward the region of low μ. Also if two oxygen solutions are in equilibrium, μ_{O_2} for the two solutions are equal.

Thus a gradient of μ_{O_2} causes a flow of molecular oxygen at a rate proportional to this gradient. That is

$$\nu_{O_2,x} = k_{O_2}\left(\frac{\partial \mu_{O_2}}{\partial x}\right) = k'_{O_2}\, RT \left(\frac{\partial \ln a_{O_2}}{\partial x}\right) \qquad \text{(E-3)}$$

where: $\nu_{O_2,x}$ = Velocity of molecules of component O_2 in direction of decreasing x, cm/sec

x = Distance, cm

k'_{O_2} = Velocity acquired by component O_2 when acted on by unit force per molecule, $\frac{\text{cm mole}}{\text{sec dyne}}$

a_{O_2} = Activity of molecular oxygen, M/liter.

Intensive parameters such as the temperature and the chemical potential can be used to provide a phenomenological description of the transport of heat and mass in systems departing from thermodynamic equilibrium. Several phenomenological laws describe such nonequilibrium phenomena in the form of proportionalities, e.g., Fourier's Law between heat flow and temperature gradient, Fick's Law between flow of matter of a component in a mixture and its concentration gradient, Ohm's Law between electrical current and potential gradient, Newton's Law between shearing force and velocity gradient, and the chemical reaction law between reaction rate and chemical potentials.

Some of these phenomenological laws have been used to describe a variety of dynamic systems occurring in natural and waste waters. In such complicated systems, however, cross-phenomena that occur may give rise to new effects. Well-known examples of this are the coupling of diffusion and heat conduction giving rise to thermal diffusion, which is also called the "Soret Effect," and its inverse phenomenon, the "Dufour Effect" (temperature difference arising when a concentration gradient exists). An exact description of such complicated systems can be achieved by the use of irreversible thermodynamics. Although such a description will define the problem confronting the investigator in such systems, it does not provide information concerning underlying mechanisms on the molecular level. Nevertheless, any acceptable specific mechanism must be consistent with its general requirements and the use of irreversible thermodynamics has been proven to be valuable in providing a phenomenological description of the system.